REGULATION OF SPOROPHORE DIFFERENTIATION IN SOME MACROMYCETES, PARTICULARLY IN COPRINI: AN OVERVIEW OF SOME EXPERIMENTAL STUDIES, FROM FRUITING INITIATION TO SPOROGENESIS *

by G. MANACHERE **

ABSTRACT - Differentiation and morphogenesis of fungi are considered as model systems with regard to the general biology of organisms. That is particularly true at the level of vegetative structures (hyphal differentiation, conidiation...) where observations and experiments can be conduced at the cellular level.
In differentiation of sporophores of macromycetes constitutes also a fundamental problem, the complexity of organized structures formed by such fungi is much greater than in the case of most popular models generally studied.
Schizophyllum commune, Fammulina velutipes and Coprini are good models for the study of differentiation and morphogenesis of macromycetes. Schematically, when "maturity of fruiting" is reached by vegetative mycelia, successive phases can be distinguished in the formation of sporophores; and correlatively, studied from specific physiological points of view: firstly, an initiation phase of fruiting release; secondly; a morphogenetic phase s.s., including the differentiation of the hymenium, and achieved by the fundamental subphases of sporogenesis and sporulation.
Numerous studies have been conduced on the influence of external factors - chemical and physical - on such successive fruiting phases of Coprini; biochemical informations are fragmentary. An overview of experimental studies about Coprini leads to a survey of some actual problems to solve when studying sporophore differentiation of macromycetes.

RÉSUMÉ - La différenciation et la morphogenèse des champignons sont considérées comme des systèmes modèles dans le cadre de la biologie des organismes en général. Ceci est particulièrement vrai à l'échelle des structures végétatives (différenciation hyphale, conidiation...) où observations et expérimentations peuvent être conduites à l'échelle cellulaire. Si la différenciation des carpophores de macromycètes constitue aussi un problème fondamental, la complexité des structures organisées correspondantes est supérieure à celle des modèles évoqués et généralement étudiés de manière privilégiée. *Schizophyllum commune, Flammulina velutipes* et divers Coprins constituent de bons modèles pour l'étude de la différenciation et de la morphogenèse des macromycètes. Schématiquement, une fois atteint par les mycéliums végétatifs un stade de "maturité de fructification", il est possible de distinguer des phases successives dans la formation des carpophores et, corrélativement, d'étudier ces phases quant à leurs spécificités physiologiques: d'abord une phase d'induction

---

* Invited paper presented at the 4th International Fungal Spore Conference, 1st July 1987. Stirling, Scotland.

** Université Lyon I - Claude Bernard, Laboratoire de Différenciation fongique, U.A. Mycologie, CNRS 1127, 69622 Villeurbanne Cedex, France.

Source : MNHN, Paris

fructifère; ensuite une phase morphogénétique s.s., incluant la différenciation de l'hyménium et s'achevant par les sous-phases fondamentales de sporogenèse et de sporulation.

De nombreux travaux ont été conduits sur l'influence de facteurs externes - chimiques et physiques - sur les phases fructifères successives de Coprins; les informations biochimiques sont fragmentaires. Une revue de travaux expérimentaux sur les Coprins permet d'envisager quelques problèmes actuels à résoudre et relatifs à la différenciation des sporophores de macromycètes.

KEY WORDS : sporophores, Macromycetes, Coprini, fruitbodies, fruiting initiation, photomorphogenesis, meiosis, sporogenesis, biological rythms.

"Ultimately, chemostructural differentiation of the several types of functional hyphae shaping the elaboration of reproductive stromas in the higher Ascomycetes and Basidiomycetes raises the most provoking and mostly unanswered questions pertaining to the visual achievement of macrofungal morphogenesis in nature." (TURIAN, 1983).

Differentiation and morphogenesis of fungi are considered as model systems with regard to the general biology of plants and, eventually, of others organisms. That is particularly true at the level of vegetative structures (hyphal differentiation, conidiation,...) where observations and experimentation can be conduced at the cellular level. Such systems are eventually genetically controlled and amenable to biochemical researches (cf. various papers in SMITH, 1983; LOVETT, 1985).

If differentiation of sporophores of macromycetes constitutes also a fundamental problem, it appears that the complexity of organised structures formed by such fungi is much greater than in the case of most popular models generally studied for: ".. the development of any organized fungal structure requires that hyphae grow toward one another and cooperate in formation of the differentiating organ; this is the diametrically reversed character of the invasive undifferentiated mycelium. We are almost totally ignorant of the factors which control this peculiar reversal in behaviour" (REIJNDERS & MOORE, 1985). Indeed the influences of environmental - nutritional and physical - factors on primordia initiation and general morphogenesis of sporophores are rather well known (MANACHERE, 1978, 1980, 1985), but the biochemical events really induced and regulated by such factors are unknown in most cases. The most precise results concern actually *Schizophyllum commune* (cf. various papers, in SCHWALB & MILES, 1978; WESSELS, 1985). In a such model species, general biochemical processes were described in parallel with fruiting evolution and according to genetical characteristics (WESSELS, 1978, 1985). Coprini are also considered as good models for the studies of differentiation and morphogenesis of higher fungi (MANACHERE, 1970, 1974; MOORE & al., 1979; MOORE, 1984a, b) particularly from a photobiological point of view. It is actually interesting to notice that the principal characteristics of photoinduction of fruiting and photomorphogenesis of sporophores - including abortion of primordia under uninterrupted light, described and precisely defined for *Coprinus congregatus* (MANACHERE, 1961, 1970, 1971) were recently described for *Coprinus cinereus* (BALLOU & HOLTON, 1985). That appears as a clear confirmation of the general value of our model. Nevertheless, for Coprini, biochemical information remains fragmentary and essential problems remain to be solved.

A true difficulty is that final differentiation of sporophores of macromycetes results from the progressive integration of successive differentiation phases. Such

Source : MNHN, Paris

interdependant phases are often studied by different searchers acting - and thinking ! - as "specialists" rather than as "generalists". This paper will try to give ... the "hybrid point of view of a ... specialised generalist"!

The initiation and subsequent development of carpophores from a vegetative mycelium depend firstly, of course, of the genetical ability to fruit of such a mycelium (cf. MEINHARDT & ESSER, 1983; RAPER, 1983; CASSELTON & ECONOMOU, 1985). They depend also of an adequate preliminary vegetative growth: physiological conditions necessary to obtain "maturity of fruiting" at the level of a fungal thallus remain to be defined for a majority of species. Of course, in most cases mycelia capable of producing normal carpophores result from the plasmogamy of two haploid mycelia, genetically compatible; according to a scheme of either bipolarity or tetrapolarity; but, in reality, there are many variants which complicate these simple modes (KÜHNER, 1977). It appears that a majority of studies have been conducted on the influence of external factors - chemical and physical - on successive fruiting phases of some basidiomycetes. Schematically, when "maturity of fruiting" has been reached by vegetative mycelia; two successive phases can be theoretically distinguished in the formation of basidiomycetes carpophores: firstly an initiation phase and fruiting release; secondly a morphogenetic phase including the differentiation of the hymenium and achieved by the fundamental subphase of sporogenesis, itself ended by sporulation.

INITIATION PHASE

The accomplishment of this phase is evidently simultaneously dependent on various environmental factors and more or less well defined endogenous factors (genetical and physiological factors) (reviewed in MANACHERE, 1978, 1980). Only some fundamental aspects will be discussed here.

Photo-induction of fruiting

Initiation phase of fruiting is a process of fungal differentiation often photo-controlled, particularly in the cases of various Coprini. For instance, illuminated cultures of *Coprinus congregatus* produced a ring of primordia on a minimal liquid medium; just behind the front of growing hyphae at the time of illumination, and no subsequent part of mycelia could be induced (DURAND, 1983b, Fig. 1). ROSS (1982, 1985) also reported the existence of an area of inducibility in a "pale mushroom" phenotype of the same species, but no primordia developed on solid medium until the mycelia had reached the edge of the plate. Similar results have also been reported for *S. commune* (RAUDASKOSKI & YLI-MATTILA, 1985). In the ROSS' experiments, photoinduction appears to be "memorized" until vegetative growth is achieved. Yet, we have demonstrated that photo-induction can be also "memorized" by fully developed mycelia (MANACHERE & BASTOUILL - DESCOLLONGES, 1985, Fig. 2). So, fruiting in dark-grown cultures of *C. congregatus* ("dark mushroom" phenotype, usually studied in our laboratory) is induced by a 12 h light-break. After a few days, the majority of cultures present characteristic etiolated primordia. Such primordia can show a normal development if submitted to suitable photoperiodic regime before abortion. Moreover, in some cases, it was established that photo-induction is "memorized" without recognizable morphological effects, for a period of 8.5 to 9.5 days. In parallel, the visible etiolated photoinduced primordia abort after about 4.5 to 7.5 days in darkness.

Source : MNHN, Paris

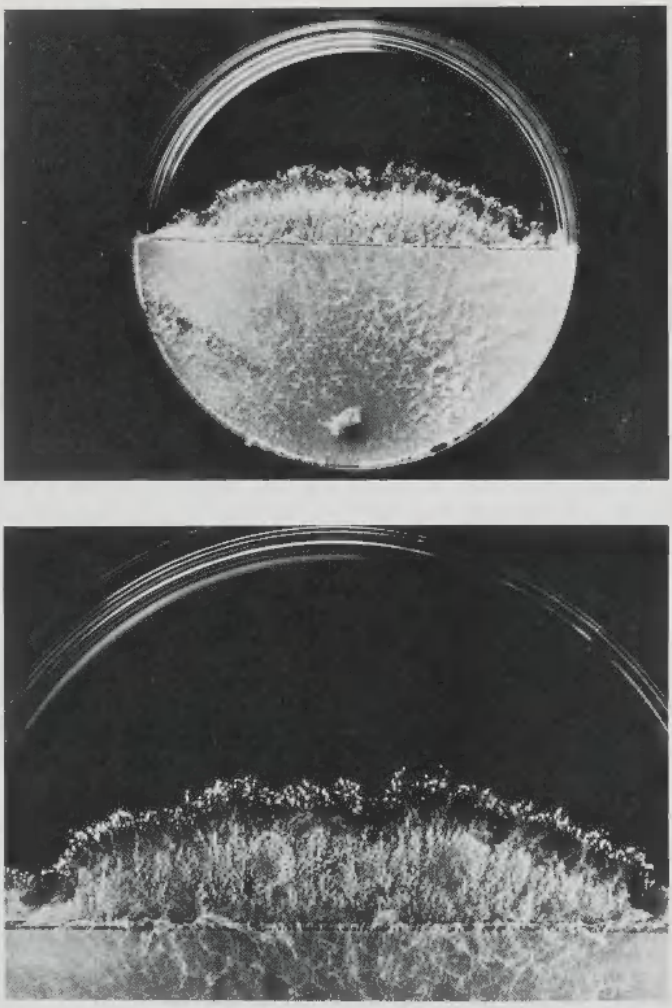


Fig. 1 - Localization of light-induced primordial formation of *Coprinus congregatus* (after DURAND, 1983b).
The fungus was cultured in partitioned plastic Petri dishes. The first compartment was filled with 20 ml of a semi-synthetic malt-agar medium. The second compartment received a salt liquid medium. After inoculation of the solid medium with a mycelial plug from stock cultures, test cultures were placed in continuous darkness at 25°C. Cultures grown in darkness for 9 days were then exposed to a 24h white light period. As a result of photo-induction, cultures produced a ring of primordia on liquid medium. Photograph was taken at the end of the light period. Dish diameter: 9 cm.
N.B.: in full darkness, cultures produced sclerotia and no fruiting primordia.

The action spectra for the initiation phase of *C. congregatus* showed peaks of effectiveness at 260, 280, 370 and 440 nm (DURAND & FURUYA, 1985, Fig.

Source : MNHN, Paris

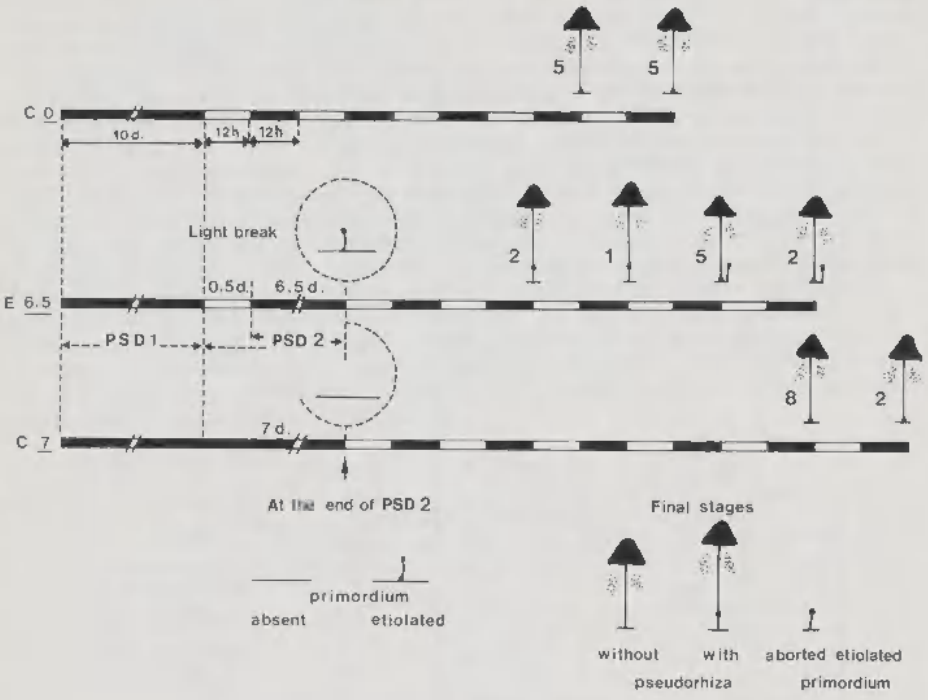


Fig. 2 - "Memorization" of photo-induction in *Coprinus congregatus* (after MANACHERE & BASTOUILL - DESCOLLONGES, 1985).
Experiments were conducted to determine the effect of short irradiations (for instance 12h ■ 300 mWm^{-2} , white light) on the initiation and differentiation of sporophore primordia and on the morphogenesis in darkness of such photo-induced primordia. Photo-induction is characterized either by development in darkness of etiolated primordia, or by ■ subthreshold state where no visible primordia are recognizable, but where interaction with a subsequent light pulse can be observed. The photo-induced primordia abort after about 4,5 to 7,5 days in darkness. The photo-induction itself is memorized for ■ longer period (8,5 to 9,5 days) in darkness.
N.B.: the figure indicates the fruiting pattern of typical experimental treatment (E) and corresponding control cultures (C0, C7). The experimental procedure is represented below. Size of primordia and final sporophores are not to scale. P.S.D.1 and P.S.D.2 = successive pre-stays of cultures in darkness following inoculation; the cultures (10/series) are then submitted to (12h, 12h) light regime.

3). A pratically identical action spectrum for fruit-body formation of *Schizophyllum commune* was also established (YLI-MATTILA, 1985). The nature of the hypothetical "cryptochrome(s)" photoreceptor(s) is unknown.
Nevertheless, according to the more recent action spectra, it seems to be flavin photoreceptors rather than carotenoids (DURAND & FURUYA, 1985; DU-

Source : MNHN, Paris

Inhibitor (M)		Primordial production	Sclerotial production
Phenylacetic acid	10^{-2}	26	-
	5.10^{-3}	50	25
	10^{-3}	82	52
	5.10^{-4}	82	88
NaN_3	10^{-3}	40	0
	10^{-4}	75	38
	10^{-5}	76	70
	10^{-6}	105	101
KI	10^{-2}	102	-
	5.10^{-3}	107	-
	10^{-3}	100	-
5-Fluorouracil	5.10^{-5}	[illegible]	1
	10^{-5}	26	35
	5.10^{-6}	80	59
	10^{-6}	92	78
	5.10^{-7}	110	85
Cycloheximide	5.10^{-5}	0	6
	$2.5.10^{-5}$	15	-
	10^{-5}	103	24
	5.10^{-6}	-	54
	10^{-6}	110	94

Table 1 - Effects of various inhibitors on primordial and sclerotial formation in *C. congregatus* (after DURAND, 1983b, 1987).
Inhibitors were applied for 3.5h during primordial photoinduction (3h of blue light) and for 6h during chemically induced sclerotial formation in darkness. Inhibitor solutions were added to the liquid salt medim (cf. Fig. 1, and more details in DURAND, 1983b). The results show potassium iodide did not inhibit blue protoinduced promordial formation. Phenylacetic acid and sodium azide inhibited the primordial and sclerotial formation. The activity of phenylacetic acid on the non photoinduced process of sclerotial formation in *C. congregatus* leads to the conclusions that this drug does not act at the photoreceptor level.
N.B.: primordial and sclerotial productions are expressed as percentage of the controls without inhibitors.

RAND, 1985, 1987). DURAND'special culturing procedure on liquid medium offers major advantages: particularly the precise localisation of a predictable area of differentiation of primordia of *C. congregatus*, and also the possibility for applying and removing chemicals to the growing mycelia at any time before or after photoinduction. But experiments using inhibitors known to react with illumi-

Source : MNHN, Paris

nated flavins (potassium iodide; phenylacetic acid...) demonstrated that these inhibitors do not act - while having other effects - at the photoreceptor level and can no longer be regarded as specific inhibitors of this blue-U.V. light response (DURAND, 1985, Table 1). According to DURAND (1987), the most promising approach to elucidate the nature of "cryptochrome" is to use a system in which the photoreceptor level can be perturbated. Experiments are being carried out to isolate riboflavin requiring mutants in *Coprinus congregatus*. So, biochemical and spectroscopic analysis of such strains should lead not only to elucidation of "cryptochrome" and of the primary events of the phototransduction chain but, also, to a better comprehension of fruiting initiation from a general point of view. For instance, the light-induced primordia formation of *Coprinus congregatus* was inhibited by various inhibitors of nucleic acid and protein synthesis (DURAND, 1983b, Table 2). Cycloheximide and a phenylalanine analogue (4 fluophenylalanine) prevented the formation of primordia, and the incorporation of leucine during photoinduction was substantially reduced in the presence of cycloheximide. Results suggest that cycloheximide inhibited protein synthesis but not the incorporation of precursors of nucleic acid synthesis. Actual results suggest the involvement of RNA and protein synthesis in light-induced primordial formation in *C. congregatus*.

Inhibitor	Before photoinduction	During photoinduction	After photoinduction
5-Fluorouracil (10^{-5}M)	77.9	26	85.4
Cycloheximide (10^{-5}M)	103.6	102.7	44.7

Table 2 - Effect of time application of inhibitors on photoinduced primordial formation of *C. congregatus* (after DURAND, 1983b).
The duration of inhibitor application was 3.5h and the length of light-induction (blue light) period was 3h. It appears that fluorouracil inhibited to a greater extent primordial formation when added in liquid salt medium (cf. Fig. 1) during the photoinduction period, whereas cycloheximide inhibited primordial formation only when added after the photoinduction period. These results presented evidence for the requirement of RNA synthesis *de novo* followed by protein synthesis in primordial photoinduction.
N.B.: primordial formation is expressed as percentage of the controls without inhibitors.

Gene expression and induction of fruiting

More generally, if actually induction of fruiting of numerous lower and higher fungi is rather easily controlled, very little is known about the mechanisms of transduction of external stimuli into a differentiation response such as fruiting initiation, and of course, following events (sporophore formation,... sporogenesis...). Studying gene expression during basidiocarp formation in *Schizophyllum commune*, WESSELS and co-workers (cf. WESSELS, 1985) detected "... few differences in proteins between monokaryons and the derived dikaryon. Furthermore, it was observed a regulation of RNA sequences... confined to the period of

Source : MNHN, Paris

basidiocarp formation in the dikaryon. The regulation involved a limited number of abundant mRNAs for a number of which cloned cDNA sequences were obtained and organized". By molecular cloning of RNAs differentially expressed in monokaryons and dikaryons of *S. commune*, a role of the detected dikaryon-specific RNAs in fruiting was recently suggested by MULDER & WESSELS (1986). Between several observations, it was noticed that nine different RNAs abundantly present in the fruiting dikaryon are present in very low or undetectable levels in vegetative growing monokaryons or the dikaryon. In spite of major difficulties, such studies about gene expression during basidiocarp formation have to be developed not only in the particular case of *S. commune* but also in the case of more classical basidiomycetes, particularly Coprini. Indeed, morphogenesis of such agaricales is more complex than morphogenesis of *S. commune*. Nevertheless it would be useful to study gene expression during basidiocarp formation in Coprini not only by a comparison of monokaryons and the derived dikaryon, but also by a comparison of dikaryons induced and non-induced to fruiting (photo-induced or not, for instance) or by a comparison of strains from the same species but more or less sensitive to light.

The problem of "fruiting-inducing substances"

For several decennies, monokaryotic fruiting has been observed in the case of some normally heterothallic species (cf. STAHL & ESSER, 1976) and also in monokaryotic cultures when wounded (ex: *S. commune*, RAPER & KRONGELB, 1958; LEONARD & DICK, 1973): models relating the various genes implicated in monokaryotic sporophore production in *S. commune* have been proposed and evaluated on their ability to explain the observed data (LESLIE & LEONARD, 1979). At last, "abnormal" monokaryotic fruiting can equally be induced from a monokaryotic mycelium of *Coprinus macrorhizus (alias C. cinereus* according to VERRINDER GIBBINS & LU, 1984) by acellular extracts of carpophores of a dikaryotic mycelia of this mushroom (UNO & ISHIKAWA, 1971). An identical effect can be obtained with acellular extracts of other basidiomycetes (e.g. *Lentinus edodes, Pleurotus ostreatus, Flammulina velutipes...)*. Carpophores produced in these conditions are not as well developed as basidiocarps produced by dikaryotic mycelia: nevertheless, the basidiospores which they form are viable, germinate, and are of the parental type. EGER (1965a, b, 1968) indicated that the production of carpophores from mycelia of *Flammulina velutipes* can be stimulated by the addition of basidiocarp fragments of this species or other species. This is the case for *Pleurotus "Florida"*. UNO & ISHIKAWA (1971) attributed the inducing power of extracts to a fruiting-inducing substance (F.I.S.). They identified this as cyclic AMP and/or cAMP-binding proteins (UNO & ISHIKAWA, 1973, 1976, 1982). However, SCHWALB (1974) with *Schizophyllum commune*, WOOD (1976, 1979) with *Agaricus bisporus* and von NETZER (1977) with *Pleurotus ostreatus* could not find any evidence that cAMP would acts as a fruiting inducer. Conversely, a "F.I.S. analog" was isolated from *A. bisporus* sporophores and this induced haploid fruit-body formation in *S. commune* (RUSMIN & LEONARD, 1975, 1978): it does not appear to be cAMP, but it may be a peptide. Some of these observations are comparable to others (reviewed by MANACHERE, 1980) showing the stimulating fruiting power of carpophore fragments placed on a vegetative mycelium (for instance *C. congregatus*, MANACHERE, 1976, 1977; ROBERT, 1978). Stimulating substances do not appear to be specific (cf. EGER, 1965b, 1968).

In parallel, there are also examples of microorganisms stimulating the fruiting of higher fungi. For instance, SALAS & HANCOCK (1972) pointed out that

Source : MNHN, Paris

Penicillium oxalicum brings about a spectacular fruiting initiation and basidiocarp production in *Mycena citricolor* when the two species are co-cultivated. One or more unidentified basidiocarp-stimulating-substances (B.S.S.) were involved. Mushroom growers consider actually that some more or less specific microorganism have a positive effect on the release of fruiting of cultivated fungi, particularly *Agaricus bisporus* (COUVY, 1973). WOOD (1976) thought that such microorganisms do not release fruiting-stimulating metabolites but that they contribute to the suppression of metabolites produced by the *Agaricus* vegetative mycelium which could prevent fruiting. This last hypothesis is considered the most plausible, as fruiting was not stimulated by culture filtrates not by suspensions of *Pseudomonas putida* or other bacteria generally present in various *Agaricus* cultures. Moreover, fruiting of such mushrooms in axenic cultures is promoted by the presence of active charcoal (EGER, 1961; COUVY, 1974; LONG & JACOBS, 1974). Thus, the role of the microflora accompanying *A. bisporus* mycelium might to be to eliminate fruiting inhibitors... rather than to produced inducing compounds !

Finally, it seems that, as for "florigen" for higher plants, the hypothetical substance(s) supposed to cause floral initiation (cf. WILKINS, 1969), F.I.S. remain "a physiological concept rather than a chemical reality". It seems also that the concept of F.I.S. covers a large spectra of molecules having a more or less defined role in general metabolism rather than a specific power in reproductive processes in fungi (ex: cerebrosides and ceramides from mycelia of *S. commune* acting as F.I.S. on the same species, KAWAI & IKEDA, 1982; sphingolipids from wheat grain acting also as F.I.S. on *S. commune* , KAWAI & al., 1986; anthranilic acid from a strain of Actinomycetess inducing the formation of the stipe of the fruiting bodies of *Favolus arcularius* "even under dark conditions", MURAO & al., 1984; ... or ammonia inducing fruiting of *Coprinus cinereus* in darkness, MORIMOTO & al., 1981, etc.).

As mentioned above; monokaryotic fruit-bodies are generally more or less abnormalous. Nevertheless, recently, it was observed that originally monokaryotic cultures of *Coprinus cinereus* developed normal fruiting bodies when subjected to particular nutritional stress from periods ranging from 3 weeks to several months (VERRINDER GIBBINS & LU, 1984). The fruiting bodies arose on dikaryotic tissue complet with clamps connexions, and underwent a normal meiosis with four spores resulting from each basidium. IT appears, in this particular case, that a stress can induce a rearrangement of the genome at the level of incompatibility factors. The mechanism for such a transformation of monokaryotic into dikaryotic tissue without mating is actually difficult to explain.

Some metabolic aspects of fruiting initiation

Nutritional researches on fruiting initiation are not reviewed here (MANACHERE, 1980). Nevertheless, such researches naturally lead to the problem of metabolic bases of the phenomenon in basidiomycetes. Fragmentary data are known relative essentially to *Agaricus bisporus, Schizophyllum commune, Coprinus cinereus (alias "lagopus")* and *Coprinus congregatus*, but practically, they do not allow us to suggest precisely those pathways implicated in initiation of fruiting. Metabolic bases of the initiation phase of fruiting of macromycetes (particularly glucose effect on fruiting) were reviewed (MANACHERE & al., 1983). Here will be evocated only some fundamental results.

When studying phenoloxidase activities in relation to fruiting of *A. bisporus*, TURNER (1974) showed that laccase activity characterizes the vegetative phase,

Source : MNHN, Paris

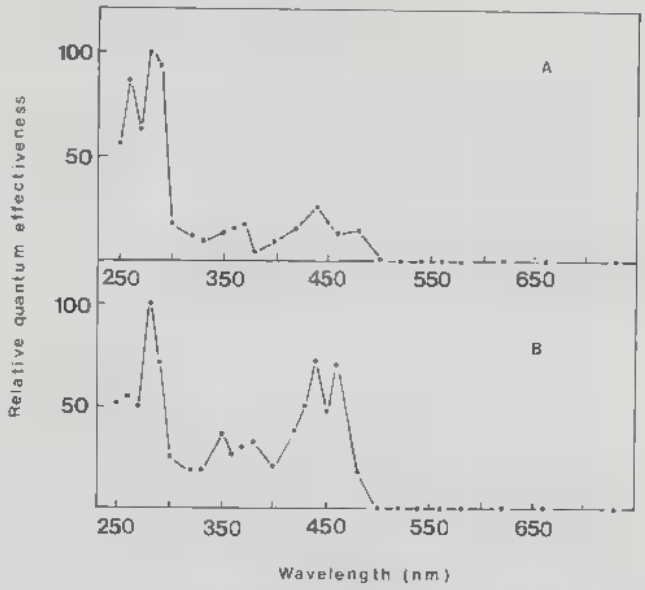


Fig. 3 - Action spectra in fruiting of *Coprinus congregatus* (after DURAND & FURUYA, 1985).
a) primordial photoinduction; b) photo-inhibition of development of "dark sensitive primordia" (cf. Fig. 4). The ordinate value for the most effective wavelength was set at 100.

whereas tyrosinase activity increases at the time of initiation and development of sporophore buds. TURNER considers that the increase of extracellular laccase in the compost (usual substratum of *A. bisporus* formed by fermented straw) during the colonization phase, and then its disappearance at the time when the first carpophore flush is developed, is connected to an attack of the substrate lignin by the mycelium during the course of its growth. Activation of tyrosinase during the formation of reproductive structures could results from the impoverishment of the medium at the end of the vegetative phase. WOOD & GOODENOUGH (1977) observed with the same mushroom in axenic cultures an inverse relation between the activity of carboxymethyl-cellulase and laccase as fructification progressively advances; they did not note any significant variation in the activity of other enzymes (xylanase, laminarase, acid and alkaline phosphatases, proteases). An increase of cellulase could contribute to the liberation of sugars which can be assimilated by the mycelia and therefore participate in carpophore formation. It can be noticed that, recently, DE VRIES & al. (1986) have observed that "... growth at 30°C in darkness prevented fruit-body formation of *Schizophyllum commune* and induced the dikaryon, but not the progenitor monokaryons, to excrete laccase into the medium in amounts up to 3% of the extracellular proteins. The competence of the dikaryon of the same species to produce laccase activity, in contrast to the component monokaryons, was also noted

Source : MNHN, Paris

by LEONARD & PHILLIPS (1973)". A major difference appears with observations on *A. bisporus* : the decline in laccase activity from dikaryon of *S. commune* occurred in the absence of fruit-body formation, after glucose exhaustion in the medium, and on the opposite, extracellular laccase activity remained practically unvariable, at a maximal level, during numerous successive days, when non-fruiting mycelia of *A. bisporus* were submitted to analysis (WOOD & GOODENOUGH, 1977).

Although they are fragmentary, other results show that the passage from the vegetative phase of mycelial growth to the initiation phase of primordia is accompanied by important metabolic changes. WESSELS (1965) showed that the metabolic activity of a whole culture of *S. commune*, as determined by gaseous exchanges, is firstly fermentative and, when the initiation phase of primordia commences, becomes oxidative. WESSELS thus established for *S. commune* a pattern which seems of general relevance by those who have studied the metabolic aspects of fungal reproduction (cf. TURIAN, 1969). In fact, this metabolic change appears to be related to variations in amounts of various metabolites, particularly carbohydrates, firstly in the medium and later in the vegetative mycelium. Thus, WESSELS showed that fruiting initiation of *S. commune* can only take place if exogenous carbon and nitrogen sources are available, but, over and above the initial stage, these requirements can differ. STEWART & MOORE (1974) showed that the quantity of reducing sugar and nitrogen in the form of α-amines in the medium diminished between the beginning of the culture and the observation of the first carpophore buds of *Coprinus lagopus*. ROBERT (1977b) showed that the fruiting initiation of *C. congregatus* occurs when glucose from the medium has been almost totally removed. Generally the pH medium of the culture has become more alkaline when primordia appear (*C. lagopus*, STEWART & MOORE, 1974; *C. congregatus* ROBERT, 1977b, HORRIERE, 1977) but this may be also related to senescence of the culture (ROBERT, 1977b).

Independently from the problematic question of F.I.S. previously mentioned, one can notice recent observations of increase in the level of cAMP in relation with primordia light-induction of *Schizophyllum commune* (YLI-MATTILA, 1987): it seems possible that cAMP could control the breakdown of reserve polysaccharides; for, in the evocated studies, the sharp increase observed in the nucleotide level took place earlier than a recognized decrease of reserve polysaccharides (cf. RAUDASKOSKI & SALONEN, 1984). Experiments are to be conducted to study this hypothesis which concerns various fungi, macromycetes and others, where cAMP has been suggested to be involved in morphogenetic and developmental processes.

MORPHOGENETIC PHASE

Some metabolic aspects of morphogenesis

Metabolic aspects of the morphogenetic phase of macromycetes (particularly identity and mobilisation of reserves, enzymatic activities and metabolic pathways) were recently reviewed (MANACHERE & al., 1983). Only some fundamental results will be discussed here.

From fruiting initiation, the growth of basidiocarps implies the transfer of water and various materials from the original mycelium (*Agaricus bisporus*, BONNER & al.,1956; *Coprinus lagopus*, MADELIN, 1956a; *Polyporus brumalis*,

Source : MNHN, Paris

PLUNKETT, 1958; *Schizophyllum commune*, WESSELS, 1965). MADELIN (1960) found, at the time of release of fruiting of *C. lagopus*, that food reserves - probably glycogen and proteins - from inflated cells of the vegetative mycelium disappeared.

Once the initiation phase is accomplished, nutritional and metabolic aspects are too intimately interwined to be distinguished from each other. In order to simplify matters, only the principal species taken as examples earlier will be considered. As far as *Schizophyllum commune* is concerned, WESSELS (1965) established that the respiratory ratio is above 1 until the primordia are formed, then decreases and moves towards 1. The consumption of oxygen remains high as long as exogenous glucose has not been totally exhausted and then falls sharply with a further decrease during carpophore maturation. During growth of primordia, while exogenous glucose must be present, an exogenous nitrogen source is unused; the needs of primordia can be met from nitrogen compounds coming from the vegetative mycelium. Subsequently, during the final phase of pileus formation, there is no need for exogenous carbon or nitrogen sources; indeed, the supply of the exogenous carbon source obstructs formation of pilei. WESSELS showed that the completion of pileus formation of carpophores depends on a low but continued supply of glucose, resulting from the hydrolysis of glucan following the breakdown of arborted primordia and mycelium cell walls. In fact, on a vegetative thallus of *S. commune* or other basidiomycetes, not all carpophore buds accomplish their development, and only some buds reach the terminal stage of maturity. The few data given above show the complexity of this problem. The progression of basidiocarp morphogenesis depends on complex nutritional and metabolic interactions: medium nutrients-vegetative mycelium, vegetative mycelium-primordia and dominating primordia-dominated primordia. In the case of *C. congregatus* when studying the behaviour of cultures on liquid media; one can notice that primordia formation takes place only when the mycelial growth declines strongly, this being correlated with carbohydrate depletion of the medium. Thereafter growth of primordia is dependent only on nutrient supply from the basal medium until the fruit-bodies reach maturity: carbohydrates reserves are used during fruiting and strongly metabolized during the last 48 to 24 h of fruit-bodies maturation (i.e. during stipe elongtion and cap expansion). It has been established that a hot-water-soluble polysaccharide is particularly used during these processes (ROBERT, 1977b, 1978). Rhythmic production is succesive flushes ends with depletion of this carbohydrate reserve which constitutes the fruiting potential of the cultures.

On the other hand, basidiocarp morphogenesis is also a function of inter-relations between the stipe and the pileus. Thus, the terminal phase of carpophore development of *C. congregatus* depends, according to ROBERT (1977a), on the accumulation of carbohydrates and proteins apparently translocated from the stipe to the maturing pileus. The final stage of morphogenesis of this *Coprinus* is not however autonomous: as excised carpophores cannot reach normal size even when excision is done during the final hours of development (BRET, 1977b). Similarly, carpophores excised from *Agaricus bisporus* do not reach normal size (TURNER, 1977). However, GOODAY (1974, 1975, 1982) showed that the terminal and rapid elongation phase of *Coprinus cinereus* basidiocarps is an autonomous endotrophic process. HAMMOND & NICHOLS (1976) have shown that mannitol accumulates in basidiocarps of *Agaricus bisporus* at the same time as the level of trehalose diminishes. However, at the end of development, the quantity of mannitol also decreases in the carphores and this is converted to another material used by the mycelium or the basidiocarp. According to the observations of BORRISS (1934a), the glycogen which first accumulates in the lower and pe-

Source : MNHN, Paris

ripheral parts of *Coprinus "lagopus"* stipes progressively moves towards the upper parts to be finally utilized during basidiocarp maturation. RAO & NIEDERPRUEM (1969) noted the predominance of trehalose in the stipe of *C. "lagopus"* and glucose in the caps, but made no measurement of changes in the amounts of these components during successive stages of development. Electron microscopical observations (MATTHEWS & NIEDERPRUEM, 1973) showed that, at a juvenile stage of primordia development of the same species; polysaccharides (probably glycogen) accumulate more in the lower cells of the stipe or those cells destined to give the future hymenium. According to KITAMOTO & GRUEN (1976), the largest carpophores produced by a culture of *Flammulina velutipes* utilize, during the course of their development, not only the residual glucose from the substrate but also, partially, the constituents of the mycelium and of those small primordia which do not develop fully. Trehalose, mannitol and arabitol are produced from glycogen and transferred to the stipes and pilei of carpophores which develop to completion.

Develop. stage	Control			Experiment			Inhib.
	n	L.i.	Δ L	n	L.i.	Δ L	%
- 19 h 35	36	5.6 ± 0.2	4.8 ± 0.5	57	6.1 ± 0.2	3.8 ± 0.3	19.9
- 17 h 45	34	6.3 ± 0.2	20.8 ± 1.9	47	6.5 ± 0.2	8.6 ± 0.8	58.3
- 15 h 40	42	7.1 ± 0.2	47.1 ± 1.3	65	7.2 ± 0.2	21.9 ± 1.3	53.5
- 13 h 50	51	8.2 ± 0.2	48.9 ± 0.9	57	8.7 ± 0.2	31.9 ± 1.2	34.8

Table 3 - Role of the cap on stipe elongation of *C. congregatus*: inhibitory effect of caps from photoinhibited primordia on elongation of decapitated stipes from primordia whose maturation is induced (after ROBERT & BRET, 1987).
Development stage: hours before autolysis; control: decapitated stipes only; experiment: caps from primordia developed during 5 days in continuous light were then applied, following decapitation; n: number of treated stipes.
N.B.: other informations cf. Fig. 4.

MOORE & EWAZE (1976), state that measurement of the specific activities of representative enzymes of the pentose phosphate cycle, Embden-Meyerhof-Parnas pathway and the tricarboxylic acid cycle in *Coprinus cinereus* sporophores at different stages of the development indicates that glycolysis is the major route of carbohydrate catabolism through sporophore development. Enzymes of the pentose phosphate cycle were always at lower specific activities than the enzyme of the EMP pathway, and the activities of the pentose phosphate cycle enzymes declined drastically as development proceeded. This conflicts with the findings for *Agaricus bisporus* (LE ROUX, 1965), but the changes in some enzymes were qualitatively similar to those occurring in the development of *Schizophyllum commune* (SCHWALB, 1974).

In practice, much remains yet to be done in order to understand the metabolism associated with fruiting of higher fungi. The few reports referred to above, essentially concerning the metabolism of carbohydrates in connection with the basidiocarp morphogenesis, are evidence of this. Other examples could have

Source : MNHN, Paris

been cited. Thus, STEWART & MOORE (1974) established that GDH_{NAD} (NAD-linked glutamate dehydrogenase) activity could be a component of the normal vegetative metabolism of *Coprinus lagopus* sensu LEWIS (*C. cinereus*) but activity of GDH_{NADP} increases in parallel with certain important changes in terminal development of carpophores. The authors showed that GDH_{NAD} activity is subject to catabolite repression and derepression by urea; GDH_{NADP} activity being, inversely, subject to catabolite derepression and repression by urea. Thus GDH_{NAD} could be the enzyme normally implicated in ammonia assimilation, GDH_{NADP} being reserved of specific functions associated with metabolic alterations in connexion with the phenomena accompanying terminal development of the basidiocarp. More precisely, GDH_{NADP} specific activity increases relatively more in the tissues of the cap than in the stipe and the basidiospores where it remains low. However, such a level of GDH_{NADP} activity has not been found in the caps of numerous other species of Basidiomycetes (MOORE & AL GHARAWI, 1976).

Finally, it appears that maturation of the cap of *C. cinereus* is accompanied by a specific pattern of changes in enzyme activities and metabolite levels. The most significant changes result in amplification of activity in tricarboxylic acid cycle and the urea cycle. "The system exemplifies different sorts of regulation, from substrate level to the gene level, and is an ideal model for study of the causative events that give rise to metabolic shifts which direct differentiation processes" (MOORE, 1984a). Nevertheless, generalization of particular results remains problematic. For instance, if, in *C. cinereus*, urease was found in the stipes, its absence from the pileus was thought to account for the accumulation of urea and arginine in the pilei (EWAZE & al., 1978). On the opposite, *Flammulina velutipes* seems to lack such mechanism, the concentration of urea remaining very low in both pilei and stipes during their growth (GRUEN & WONG, 1981a).

Among the histochemical studies, KOMAGATA & OKUNISHI (1969) showed that, in *Coprinus kimurae*, the activities of cytochrome oxidase, succinic dehydrogenase and acid and alkaline phosphatases are localized at the carpophore growth zone; essentially the upper part of the stipe and the edge of the pileus. Similar observations have been made for *Polyporus brumalis* (OKUNISHI & KOMAGATA, 1975). Studies on *Favolus arcularius* have confirmed the localization of respiratory enzymes (cytochrome oxydase and succinic dehydrogenase) at the level of active growth zones which are the terminal part of the stipe; the entire pileus and the young hymenia (HORIKOSHI & al., 1973).

Regulation of fruit body morphogenesis at the level of individual sporophores

The general development of sporophores (stipe elongation, cap maturation) requires a continuous supply of water and nutrients from the vegetative mycelium during most of their growth period. The identity of the translocated nutrients has not been determined. Nevertheless, the elongation of isolated whole fruit bodies of *Flammulina velutipes* was promoted by glucose and other low molecular weight carbohydrates (GRUEN & WU, 1972).

Many experiments show that, when a factor prevents pileus development, excessive elongation of the stipe follows. In fact, complex correlations regulate the individual development of the two parts of sporophores. HAGIMOTO & KONISHI (1959, 1960), HAGIMOTO (1963a, b) and GRUEN (1963) showed that one or more growth substances elaborated at the level of the hymenial lamellae probably control the elongation of carpophore stipes of the cultivated *Agaricus*

Source : MNHN, Paris

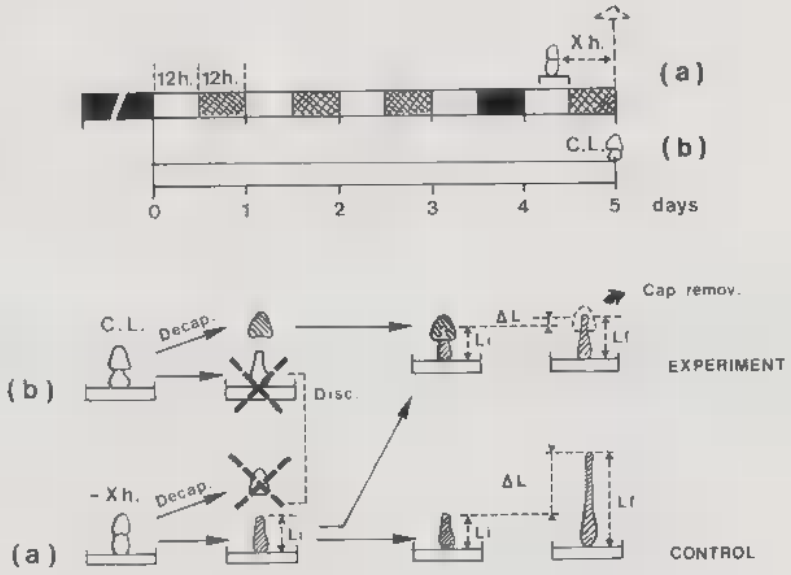


Fig. 4 - Light conditions and method used to show the presence of an inhibitor of stipe elongation in the cap of primordia of *Coprinus congregatus* developed under continuous light (original figure by ROBERT, after ROBERT & BRET, 1987).
Decap.: decapitation = beginning of the experiment; - X h: stages of development of operated primordia (cf. table 3) = X hours before autolysis (*a*); C.L.: abortive primordia produced under continuous light *(b)* ; L.i.: initial length, measured just after decapitation; L.f.: final length, measured just after a minimum of 24h in light and then, cap removal when necessary; Δ L: residual growth of only decapitated stipes (*control*) or receiving caps from C.L. primordia (*experiment*); disc.: discarded parts.
N.B.: hatched periods are periods where darkness is not needed for normal development; the black one correspond to an absolute requirement at 25°C; this last temperature was maintained during all manipulations.

so that the unilateral elimination of lamellae from a young carpophore leads to a preferential elongation at the side where the lamellae were conserved such that a curvature is produced. Similar observations have been made with *Flammulina velutipes* (GRUEN, 1976) and *Coprinus congregatus* (BRET, 1977a, b). The active substance has not yet been identified. Moreover, the cap plays an essential role in the determination of stipe elongation. So, in *A. bisporus* and *F. velutipes*, stipe elongation depends on pileus during most of the growth period; growth controlling activity originates in the lamellae (GRUEN, 1963, 1969, 1982). In *C. congregatus*, the presence of the cap is also necessary until a late stage for complete stipe elongation in developing fruit bodies (ROBERT & BRET, 1987).

It may appear that the stipe-pileus relationships are one-way, the pileus controlling the stipe elongation. This is however not the case as the stipes eventually

Source : MNHN, Paris

control the translocation of various nutrients from the vegetative mycelium to the different parts of the basidiocarps and the latter remains dependent on the original thallus until the final hours of their development.

Going from the information given above and by the analogy of our knowledge on the intervention of growth substances in tropisms of higher plants, it would seem reasonable to think that the positive phototropism (young sporophores) and negative geotropism (terminal elongation phase of mature sporophores) characterizing certain phases of basidiocarp morphogenesis could be determined by "hormones". Such hormonal substances, released asymmetrically by the hymenial lamellae of carpophores would determine the curvature of stipes by bringing out the elongation of the upper cells of the later. However, in the case of *Flammulina velutipes*, the stimulatory effect of stipe elongation by an extract of lamellae is significantly increased if it is transmitted by nutritive agar, e.g. potato dextrose agar, rather by a simple agar (GRUEN, 1976).

Little is known about the mechanism of action of lamellae on stipe elongation. *A. bisporus* lamellae produce a diffusate in agar which, when applied unilaterally, causes a stipe curvature, indicative of growth promotion (HAGIMOTO and KONISHI, 1959, 1960; GRUEN, 1967). No curvature is observed when IAA is applied instead of diffusate. In *C. congregatus*, removal of half the cap resulted in negative curvature of the stipe when the operation was performed more than 12 h before the end of the fruit body development (BRET, 1977b). Experiments consisting of joining caps and stipes of sporophores of different ages have shown the pattern of growth substances production in *C. congregatus*. Stimulation was maximal when the deposited cap came from a fruit body that was 16 h before the end of its development (BRET, 1977b). In parallel, sensibility of stipe to substances produced by hymenial lamellae was also maximal when such a stipe was 16 to 18 h before the end of its development (ROBERT & BRET, 1977, Table 3); it is noticeable that the "minus 16 h stage" is characterized by the beginning of meiotic divisions following karyogamy at 25°C (MANACHERE, 1968; MANACHERE & BASTOUILL - DESCOLLONGES, 1982). In the same species, it was possible to inhibit elongation of the stipe when this process becomes quite independent of the presence of caps (i.e. after the - 16 h stage previously mentioned): this was done by using caps from non-elongating and sporeless aborting fruit bodies obtained in continuous light. It seems that light which causes the primordia inhibition, and leads also to nuclear fusion in the young basidia (MANACHERE & BASTOUILL - DESCOLLONGES, 1982), may act by stimulating the synthesis - or only the accumulation - of an inhibitor in the cap (ROBERT & BRET, 1987, Fig. 4). In continuous light, the presence of the inhibitor appears to prevent stipe growth in the primordium, but may also directly arrest further development of meiosis.

All these results imply the production of growth- promoting or growth- inhibiting factors in the cap. However, as yet, there has been no successful biochemical characterization of the factors involved. The question is also to determine if such hypothetical factors are the same as those controlling the differentiation of the next flush, these substances being synthetized in the caps and diffusing through the stipe to the basal mycelium (ROBERT, 1982).

In some isolate examples, the control of stipe elongation by the cap appears similar to the phenomenon of apical dominance in higher plants: for instance, the removal of the cap of a young sporophore of *Asterophora parasitica* is followed, in full light, by the development of several lateral new primordia on the remaining stipe (VIOT, 1970). In darkness, mycelia of the same species produced abnormal primordia with lateral ramifications, each ended by a reduced pileus:

Source : MNHN, Paris

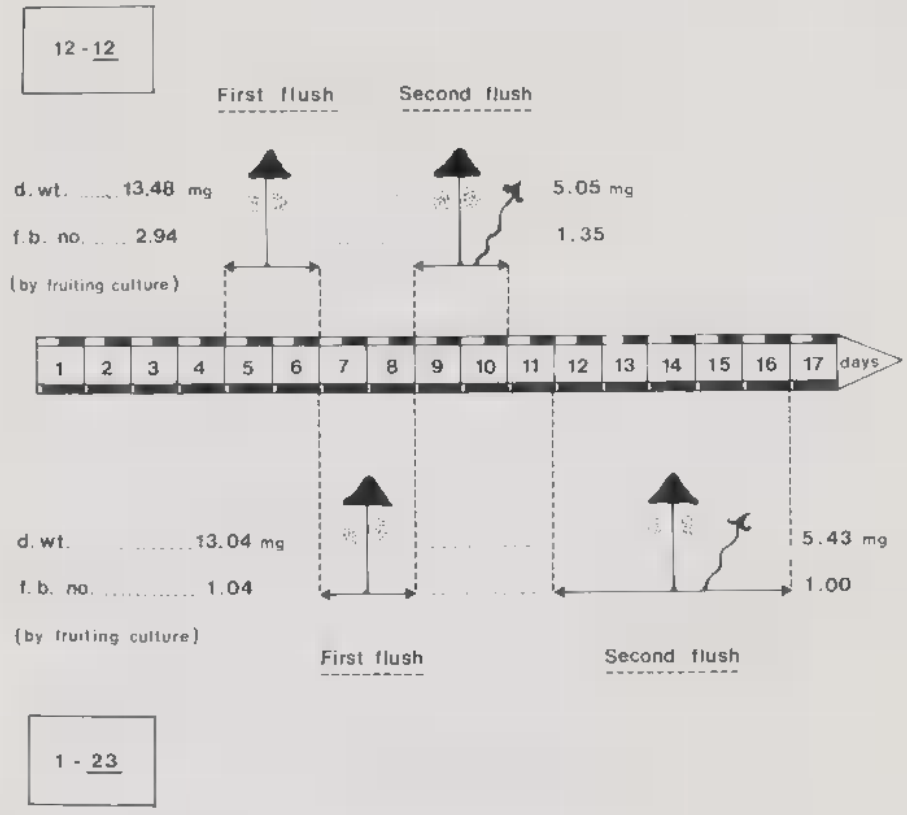


Fig. 5 - Comparison of fruiting of *Coprinus congregatus* under 2 distinct photoperiodic treatments: L D:12 12 regime (control series) and L/D: 1/23 regime (after MANACHERE & BASTOULL - DESCOLLONGES, 1983).
Number of cultures series: 25. Mean dry weight (d. wt.) and mean fruit body number (f.b. no.) by fruiting cultures and by flush are indicated. Culture temperature 25°C. Illumination radiant flux density 300 mWm^{-2} (white light).

such observations confirm previous hypothesis about physiological correlations between pilei and stipes.

Regulation of fruit body morphogenesis at the whole culture level

Periodical fruiting characterizes several higher fungi (see MANACHERE, 1980; MANACHERE & al., 1983). In the case of *C. congregatus*, when cultures are maintained for long periods under alternating daily periods of 12 h light and 12 h darkness, they produce mature sporophores according to an endogenous

Source : MNHN, Paris

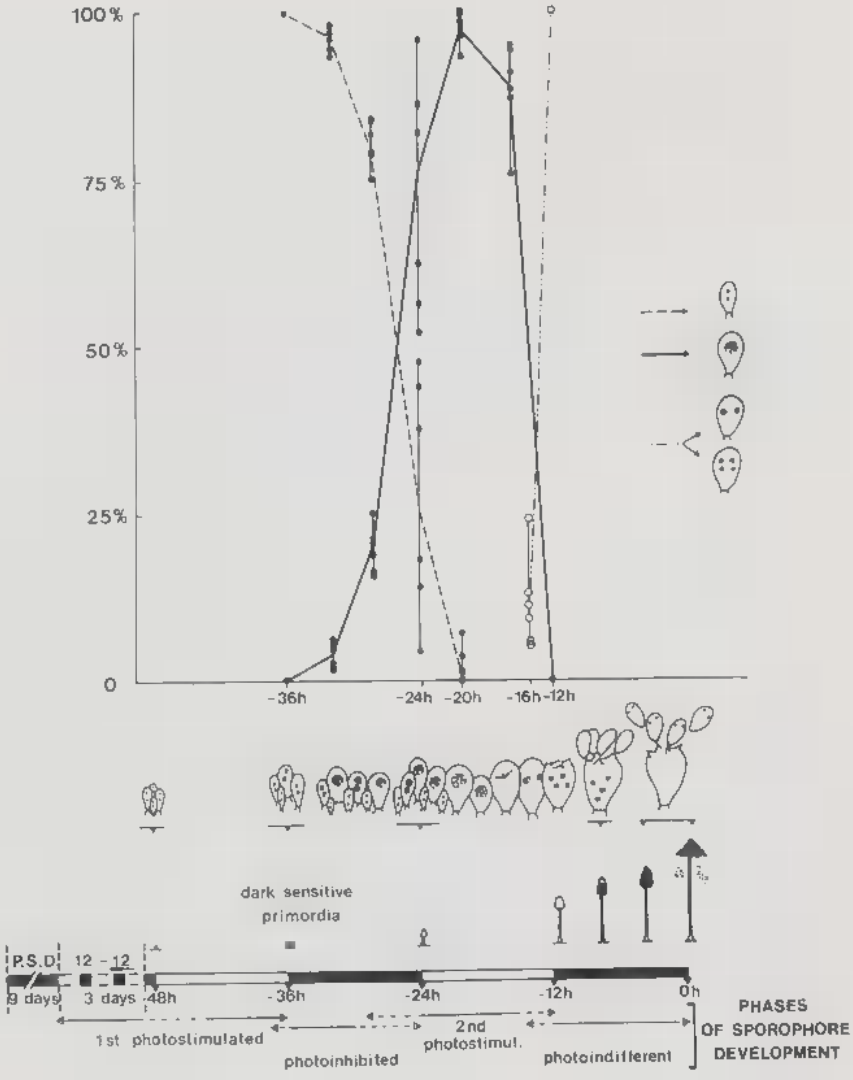


Fig. 6 - Meiosis and sporogenesis evolution in *Coprinus congregatus* under photoperiodic regime (L;D: 12.12; culture temperature: 25°C) (after MANACHERE & BASTOUILL - DESCOLLONGES, 1982). Statistical representation of the evolution of meiosis from the typical "dark sensitive stadium" till the formation of the four meiotic nuclei; behind: comparative study of cytological and morphological evolution of sporophores.

Source : MNHN, Paris

rhythm with a period which is dependent upon temperature: 4 to 5 days at a temperature of 25°C, 6 to 9 days at 15°C, for instance (ROBERT & MANACHERE, 1971; MANACHERE & ROBERT, 1972). A similar endogenous fruiting rhythm is known in the case of several other higher species, particularly the cultivated mushroom *Agaricus bisporus* (COOKE & FLEGG, 1962; WOOD & GOODENOUGH, 1977; HAMMOND, 1981).

In the case of *C. congregatus*, when mature sporophores of one or two successive flushes are picked, there is commonly a delay in the differentiation of the next flush and a decrease in - and even a suppression of - the production of this flush. Conversely, when exogenous sporophores (MANACHERE, 1977) or water extracts (ROBERT, 1978) of picked sporophores are applied, one can observe a promotive effect, i.e. an earlier differentiation of the following flush (mainly with extracts from caps) and sometimes, a more abundant production of sporophores (mainly with extracts from stipes).

When cultures of *C. congregatus* are maintained at 25°C under L D: 1 23 regime (i.e. 1 h of light per day), the first flush is delayed on the average of two days in comparison with cultures under L D: 12 12 regime and this flush generally shows only 1 fruit body culture against 3 fruit bodies culture on average under the L D 12 12 regime. Nevertheless, if dry weight is considered rather than number of fruit bodies, productivity is identical in the two series of cultures (MANACHERE & BASTOUILL - DESCOLLONGES, 1985, Fig. 5). Similar results were obtained when illumination was lowered from 300 to 10 mWm^{-2}, the cultures remaining under a L D: 12 12 regime (ROBERT, 1982). It appears that the reduction of the daily lighting (duration or energy level) is compensated by internal regulation at the level of the whole culture. Such regulation controls the global development of mature sporophores, particularly individual morphogenesis and rhythmical production. These observations confirm the physiological unity of a culture of *C. congregatus* demonstrated by previous experiments (MANACHERE, 1976, 1977). A fundamental question remains unanswered in the case of *C. congregatus* (MANACHERE & ROBERT, 1972) and all other species showing a fruiting rhythm: at what stage are the second and subsequent flushes initiated ? All the flushes may be initiated at the outset, with the later ones remaining cryptic in a stage not visible to the naked eye; or each might be initiated just before or coincident with maturation of the preceding flush.

In the case of *C. congregatus* (but also in the cases of *Sphaerobolus stellatus*, INGOLD & NAWAZ, 1967, and of *Agaricus bisporus*, COOKE & FLEGG, 1965) picking of the primordia (and not the mature sporophores as in the experiments mentioned above) leads to an acceleration of the primordia development of the following flushes, the acceleration being greater the earlier stage during which primordia are picked. This could results from nutrient deprivation of dominant primordia. Such hypothesis is coherent with past experimental demonstration of MADELIN (1956b) relative to *Coprinus lagopus* or of WESSELS (1965) relative to *Schizophyllum commune* and more recent observations of GRUEN & WONG (1981b) showing that completely developed sporophores of *Flammulina velutipes* derive their substrates indeed from the nutritive medium, but also from material stored in the rest of the colony: loss of dry weight by aborted primordia and stunted fruit-bodies parallels gains by large fruit-bodies.

Source : MNHN, Paris

Physiological aspects of sporogenesis

For several decennies, works have been conduced to determine morphological, histological and cytological characteristics of basidiocarps of numerous species of Agaricales (cf. REIJNDERS, 1963; KÜHNER, 1926 and numerous papers mentioned in REIJNDERS,1963; MOORE, 1984a, b). A particular attention has generally be given to the evolution of hymenial cells, from the

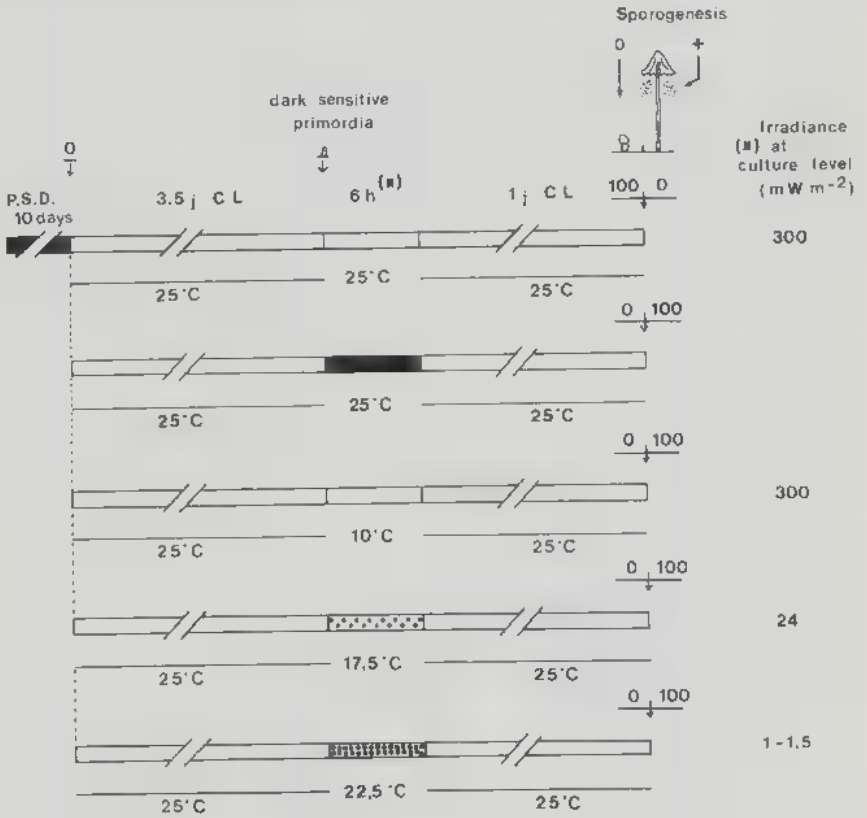


Fig. 7 - Effects of light and temperature interactions on maturation of sporophores (including sporogenesis) of *Coprinus congregatus* (after DURAND, 1983a). Cultures grown in darkness for 10 days (P.S.D. = pre-stay in darkness) were then exposed to white light (300 mWm^{-2}) without interruption during 3.5 days. At the end of this period, they have produced characteristic "dark sensitive primordia" (cf. MANACHERE, 1970, for instance). They are then submitted during 6h to various thermical and light regimen. Percentage of cultures with aborted primordia only (no basidiospores produced) and of cultures with normal sporulating sporophores is evaluated 24h after the end of the eventually maturating regimen.
N.B.: sp. +: sporophores with normal sporogenesis; sp. O: sporeless aborting primordia.

Source : MNHN, Paris

young basidia stage to the "sporulating basidia" terminal stages (cf. HUGUENEY, 1978: *Coprinus congregatus;* ROSIN & MOORE, 1985a, b: *Coprinus cinereus).* Nevertheless few data concern the physiology of sporogenesis of such macromycetes. In most cases, observations of "normal" evolution of basidia are given, including replication of DNA meiotic divisions and various ultrastructural and cytochemical aspects (LU, 1982; THIELKE, 1982; Mc LAUGHLIN, 1982). Beyond such useful reports, one can notice a lack about the control of meiotic events and of sporogenesis by external and hypothetical internal factors: in other words, the physiological aspects of basidial and basidiospore development s.s. are practically unknown.

The morphological stages and phases of development of sporophores of *C. congregatus*, and, correlatively, the nuclear behaviour of hymenial cells till complete sporogenesis are well defined and determined essentially by daily light and dark periods. Each stage is defined - at 25°C in most observations - by the number of hours before sporophore maturity, which is called 0 h stage (for instance, - 36 h stage represents 36 h before the 0 h stage). With respect to the cytological state, and particularly the development of meiosis, there is no karyogamy at the - 36 h stage. Only 50% of the basidia have diploid nuclei at the - 24 h stage. Usually, all basidia reach karyogamy between the - 20 and - 16 h stages, and the meiotic divisions begin at the - 16 h stage and are completed at the - 12 h stage (MANACHERE & BASTOUILL - DESCOLLONGES, 1982). On the other hand, at 25°C there is a photoinhibition phase between - 36 and - 24 h stages, light being necessary before and after this phase. In continuous light, after the - 36 h stage (stage of sensitivity to darkness) one can essentially observed an inhibition of stipe elongation and an arrest of meiosis at the nuclear fusion stage correlatively; one can observe no opening of pileus (MANACHERE, 1970, Fig. 6, 8).

A similar inhibitory effect of continuous light on stipe elongation and meiosis (i.e. arrest at the nuclear fusion stage) was then observed on primordia of various other species of other Coprini: "*C. lagopus*" at 35°C (LU, 1972), dikaryotic sporophores of *C. macrorhizus* at 28°C (KAMADA & al., 1978), monokaryotic sporophores of a mutant strain of the same species at 25°C (MIYAKE & al., 1980).

In *C. congregatus*, the dark requirement observed at relatively elevated temperature (20-25°C) is not found for temperatures less than 17.5°C. Normal development of primordia in continuous light (300 mWm^{-2}) was obtained by lowering the basal temperature of the culture from 25 to 10°C for 6 h (ROBERT, 1971; ROBERT & DURAND, 1979). More precisely, at low levels of irradiance (1-5 mWm^{-2}) a slight decrease in temperature (25 to 22.5°C) was sufficient to release primordia from photoinhibition (DURAND,1982, 1983a, Fig. 7). It was observed that meiosis and consecutive sporogenesis are strictly dark dependent at 25°C, but could proceed in continuous light at 10°C (Fig. 8). It appears that the dark requirement triggering meiosis and then besidiocarp maturation in several *Coprinus* species, *C. lagopus* (LU, 1972), *C. congregatus* (MANACHERE & BASTOUILL - DESCOLLONGES, 1982) could be temperature-dependent.

Furthermore, in certain higher and lower fungi the dark period which determines the terminal phase of development of sporophores, namely the phase of maturation including sporogenesis, could be replaced by lowering the temperature. For instance, some parasitic micromycetes, *Alternaria solani* and *A. tomato*, failed to develop conidia under continuous illumination at 25°C: however; the inhibiting effect of light was no longer present at 15°C (ARAGAKI, 1961; LUKENS, 1966).

Source : MNHN, Paris

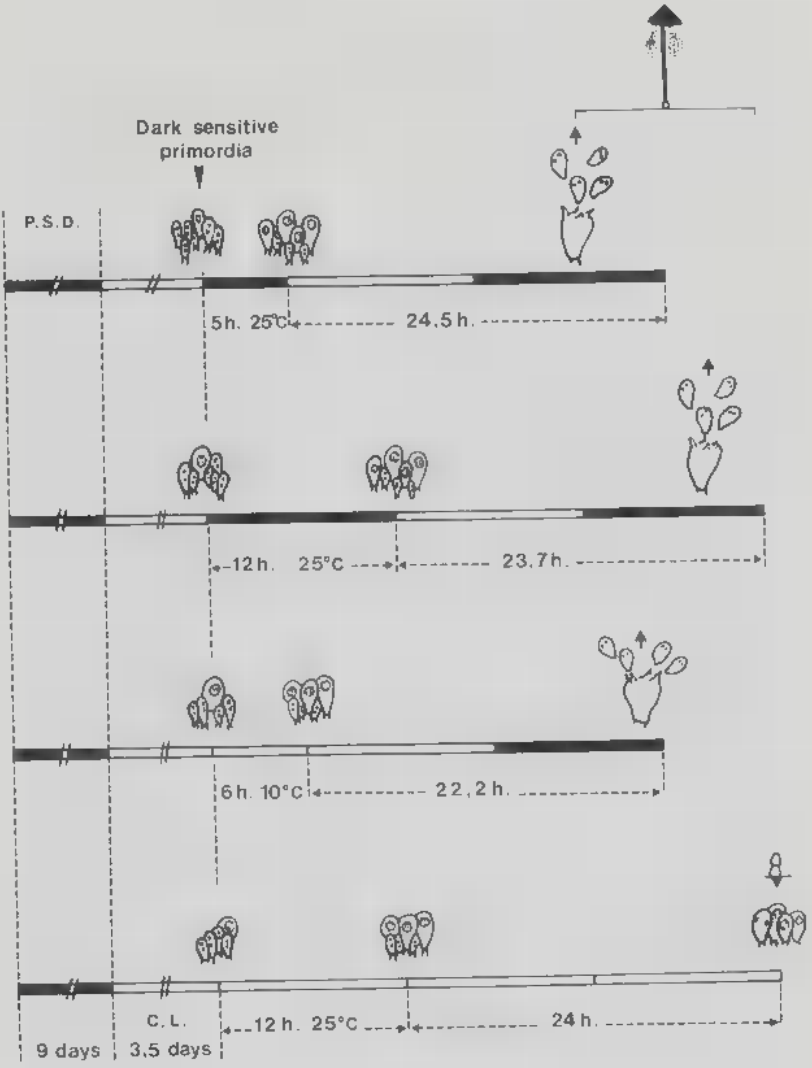


Fig. 8 - Effects of light temperature interactions on meiosis in *Coprinus congregatus* (after MANACHERE & BASTOUILL - DESCOLLONGES, 1982).
Experimental conditions: cf. Fig. 7. Primordia reach "dark sensitive stadium" after 3.5 days under continuous white light. Meiosis, and consecutive sporogenesis, is normal when such primordia are submitted to ■ dark period during 5 h, temperature: 25°C or a dark period during 12 h, temperature: 25°C or full light during 6h, temperature lowered from 25 to 10°C.

Source : MNHN, Paris

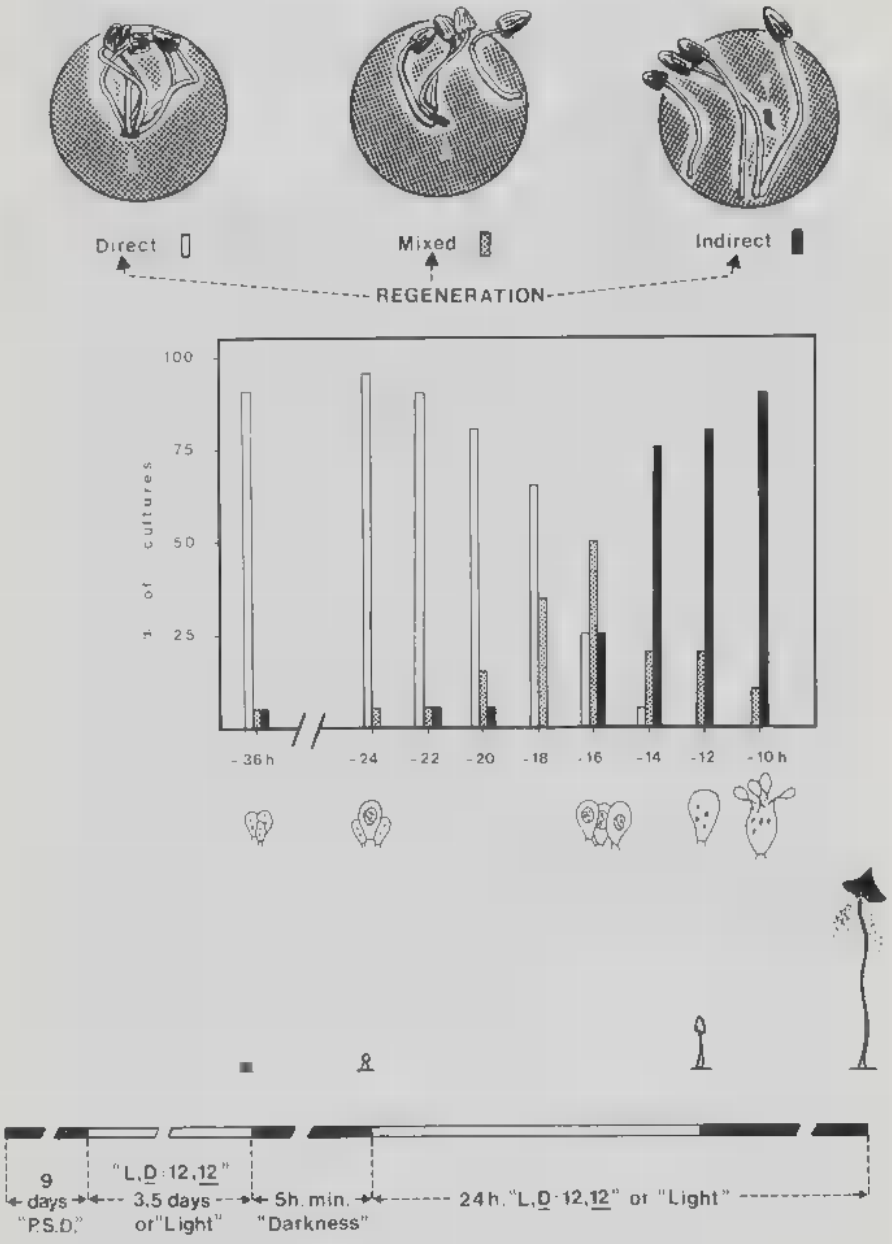
Such phenomena are observed in other groups of plants. Some of the developmental features cited for *C. congregatus* have their analogy in the photoperiodic responses of some extremely sensitive short-day plants, particularly those which can be induced by a single dark period such *Xanthium strumarium, Pharbitis nil, Lemna perpusilla* (EVANS, 1969), *Perilla ocymoides* (DERONNE & BLONDON, 1977) or strawberry (HEIDE, 1977). Therefore, floral initiation, conidia formation or fruitbody maturation can be induced by a number of alternative pathways and are not strictly dependent upon a specific photoperiodic process.

In parallel with studies on physiological correlations during morphogenesis and sporogenesis of sporophores of *C. congregatus*, recent work was conducted to determine the potential of fruiting of isolated hymenial lamellae used as inocula, at various meiotic and sporogenetic stages (BASTOUILL - DESCOLLONGES & MANACHERE, 1984, Fig. 9). When isolated lamellae were transplanted at young stages (binucleate or karyogamy stages, i.e. -36 to -24 h stages), in most cases young basidia did not develop beyond prophase I of meiosis. But inoculation was followed, practically without exception, by a direct fruiting, no sporophore of the first flush appearing on the vegetative mycelium developed around the inoculum. The first fruiting flush was observed exclusively on the inoculum. Conversely, when isolated lamellae were transplanted at older stages, meiosis and sporogenesis proceeded normally and the potential for direct renewed fruiting disappeared: the first fruiting flush was generally not observed on the inoculum but on the surrounding mycelium. These results, among others, confirm that physiological phases and stages of primordia of *C. congregatus* may be defined not only by the usual morphological characters, but also by correlated cytological stages of meiosis-sporogenesis at the level of hymenial cells.

From a biochemical point of view, one can notice interesting but isolated experiments of RAUDASKOSKI & LU (1980) studying the effects of hydroxyurea (HU) on meiosis and genetic recombination in *Coprinus "lagopus"*. The drug was applied directly at the level of hymenial cells of primordia, at various development stages. The effects of HU on the meiotic cell cycle suggest that the drug can exert its effects by inhibiting the synthesis of deoxynucleotides: the two most sensitive meiotic stages are mid-lat premeiotic S phase and pachytene-diplotene period. Recently, MOORE & al. (1987) noticed that increase in GDH_{NADP} yet observed during terminal development of sporophores of *Coprinus cinereus* (cf. STEWART & MOORE, 1974) "... was initiated as karyogamy became evident; enzyme activity stabilized for about 4 hours during meiosis, but resumed after meiosis II and continued to increase until spore maturation... It is concluded that expression of GDH_{NADP} in the fruit-body cap of *C. cinereus* is either a component part of the cellular programme involved in karyogamy, or is directly triggered by that programme. Further study of this system will be an important contribution to understanding of the immediate metabolic impact of nuclear fusion events like fertilization".

Cultures are then returned to classical photoperiodic regime (L:D: 12,12) where normal meiosis and sporogenesis are observed. One can notice that basidia remain at "nuclear fusion stage" when primordia remain under continuous light without lowering temperature.

Source : MNHN, Paris



Source : MNHN, Paris

CONCLUSION

There is an evident need for fundamental studies on different aspects of sporophore differentiation in macromycetes, from primordia initiation to achieved sporogenesis. *Coprinus* species remain useful models for such works, for instance on metabolic processes, on nature of the photoreceptors, on hypothetical fruiting-stimulating substances. The regulation of gene expression during some fundamental events, particularly during primordia initiation merits a particular attention. More than ever, the study of fungi - lower and higher - should contribute to a better understanding of various physiological general problems in plants, such as photoperiodic control of reproduction in organisms, nature of blue and U.V. photoinduced or photoinhibited reactions (cf. problem of "cryptochrome"), interactions of external factors controlling morphogenesis and sporogenesis (particularly light and temperature) endogenous rhythmical processes of differentiation of reproductive structures... In addition to such areas of prime interest, there remain diverse correlations at all phases of growth and development, both at the level of the individual sporophore and of the whole culture which constitute a physiological unit analogous to a whole higher plant.

At last, from a strictly mycological point of view, most of the physiological aspects of meiosis and consecutive sporogenesis are still unknown. Researches in this last field would indeed lead to a better understanding of the influence of various external and internal factors on the reproduction of higher fungi, when acting at the level of replication of DNA and successive meiotic events.

ACKNOWLEDGEMENTS

The author wishes to gratefully acknowledge Mrs Y. BASTOUILL - DESCOLLONGES for valuable contribution to various researches mentioned in this paper and for preparation of most figures. Thanks are also due to Mr J.C. ROBERT for his help, particularly for a critical review of this manuscript and preparation of figure 4, and to Mr R. DURAND for useful complementary indications. Furthermore, particular thanks are due to Professor G.W. GOODAY, University of Aberdeen, for a critical final review of this paper.

Fig. 9 - Influence of cytological and physiological stages of meiosis and sporogenesis on "regeneration" potential of isolated lamellae of sporophores of *Coprinus congregatus* (after BASTOUILL - DESCOLLONGES & MANACHERE, 1984).
The drawing below the abscissa represent the degree of development reached by hymenial cells when lamellae of the corresponding physiological stages (-36h, -24h, ... -10h) are used ■ inocula (20 cultures/each physiological stage).
N.B.: at the top of the figure, illustration of direct □ , mixed ⊡ and indirect ▩ regeneration.

Source : MNHN, Paris

REFERENCES

ARAGAKI M., 1961 - Radiation and temperature interaction on the sporulation of *Alternaria tomato*. *Phytopathology* 51: 803-805.

BALLOU L.R. and HOLTON R.W., 1985 - Synchronous initiation and sporulation of fruit-bodies by *Coprinus cinereus* on a defined medium. *Mycologia* 77: 103-108.

BASTOUILL - DESCOLLONGES Y. and MANACHERE G., 1984 - Photosporogenesis of *Coprinus congregatus*: correlations between the physiological age of lamellae and the development of their potential for renewed fruiting. *Physiol. Pl.* 61: 607-610.

BONNER J.T., KANE K.K. and LEVEY R.H., 1956 - Studies on the mechanics of growth in the common mushroom, *Agaricus campestris*. *Mycologia* 48: 13-19.

BORRISS H., 1934a - Beiträge zur Wachstrums und Entwicklungsphysiologie der Fruchtkörper von *Coprinus lagopus*. *Planta* 22: 28-69.

BORRISS H., 1934b - Ueber den Einfluss ausserer Faktoren auf Wachstum und Entwicklung der Fruchtkörper von *Coprinus lagopus*. *Planta* 22: 644-684.

BRET J.P., 1977a - Respective role of cap and mycelium on stipe elongation of *Coprinus congregatus*. *Trans. Brit. Mycol. Soc.* 68: 363-369.

BRET J.P., 1977b - Contrôle de l'élongation du stipe par le chapeau dans les carpophores de *Coprinus congregatus*. *Compt. Rend. 102eme Congr. Natl. Soc. Sav.* (Limoges, Avril 1977), Sect. Sci. 1: 273-284.

CASSELTON L.A. and ECONOMOU A., 1985 - Dikaryon formation. *In* MOORE D., CASSELTON L.A., WOOD D.A. & FRANKLAND J.C., *Developmental biology on higher fungi*. Cambridge University Press: 213-229.

COOKE D. and FLEGG P.B., 1962 - The relation between yield of the cultivated mushroom and stage of maturity at picking. *J. Hort. Sci.* 37: 167-174.

COOKE D. and FLEGG P.B., 1965 - The effect of stage of maturity at picking on the flushing of crops of the cultivated mushroom. *J. Hort. Sci.* 40: 207-212.

COUVY J., 1973 - Les facteurs de fructification des Agaricales et plus particulièrement de l'*Agaricus bisporus* (Lange) Sing. *Le Botaniste* 56: 103-128.

COUVY J., 1974 - La fructification d'*Agaricus bisporus* en milieu aseptique: un modèle expérimental pour l'étude des substances impliquées dans l'initiation fructifère. *Mushroom Science* 9: 157-164.

DERONNE M. and BLONDON F., 1977 - Mise en évidence chez le *Perilla ocymoides* L., plante de jours courts typique, d'un facteur de l'induction florale : les températures basses. Etudes de l'état induit acquis par la feuille en jours courts ou au froid. *Physiol. Vég.* 15: 219-237.

DE VRIES O.M.H., KOOISTRA W.H.C.F. and WESSELS J.G.H., 1986 - Formation of an extracellular laccase by a *Schizophyllum commune* dikaryon. *J. Gen. Microbiol.* 132: 2817-2826.

DURAND R., 1982 - Fruiting of *Coprinus congregatus*. Interacting effect of radiant flux density and temperature. *Experientia* 38: 341-342.

DURAND R., 1983a - Photomorphogenèse des carpophores du basidiomycète *Coprinus congregatus*. Induction de la formation et de la maturation des primordiums. Thèse Doctorat d'Etat, Université Lyon I, Villeurbanne.

DURAND R., 1983b - Effects of inhibitors of nuclei acid and protein synthesis on light-induced primordia initiation in *Coprinus congregatus*. *Trans. Brit. Mycol. Soc.* 81: 553-558.

DURAND R., 1985 - Blue U.V.-light photoreception in fungi. Review. *Physiol. Vég.* 23: 935-943.

Source : MNHN, Paris

DURAND R. and FURUYA M., 1985 - Action spectra for stimulatory and inhibitory effects of U.V. and blue light on fruit-body formation in *Coprinus congregatus*. *Pl. Cell Physiol.* 26: 1175-1183.

DURAND R., 1987 - A photosensitive system for blue /U.V. light, effects in the fungus *Coprinus*. *In:* SENGER H., *Blue light responses: phenomena and occurrence in plants and microorganisms*, vol. I, Boca Raton (Florida), C.R.C. Press Inc.: 31-41.

EGER G., 1961 - Untersuchungen über die Funktion der Deckschicht bei der Fruchtkoperbildung des Kulturchampignons, *Psalliota bispora* Lge. *Arch. Mikrobiol.* 39: 313-334.

EGER G., 1965a - Untersuchungen über die Bildung und Regeneration von Fruchtkörpern bei Hutpilzen. I. *Pleurotus "Florida"*. *Arch. Mikrobiol.* 50: 343-356.

EGER G., 1965b - Untersuchungen über die Bildung und Regeneration von Fruchtkörpern bei Hutpilzen. III. *Flammulina velutipes* Curt. ex Fr. und *Agaricus bisporus* (Lge) Sing. *Arch. Mikrobiol.* 52: 282-290.

EGER G., 1968 - Untersuchungen zur Stimulation der Fruchtkörperbildung bei einem *Pleurotus* (aus) Florida. *Theor. Appl. Genet.* 38: 23-27.

EVANS L.T., 1969 - The nature of flower induction. *In:* EVANS N., *Induction of flowering*. Ithaca, Cornell University Press: 457-480.

EWAZE J.O., MOORE D. and STEWART G.R., 1978 - Co-ordinate regulation of enzymes involved in ornithine metabolism and its relation to sporophore morphogenesis in *Coprinus cinereus*. *J. Gen. Microbiol.* 107: 343-357.

GOODAY G.W., 1974 - Control of development of excised fruit-bodies and stipes of *Coprinus cinereus*. *Trans. Brit. Mycol. Soc.* 62: 391-399.

GOODAY G.W., 1975 - The control of differentiation in fruit-bodies of *Coprinus cinereus*. *Rep. Tottori Mycol. Inst.* 12: 151-160.

GOODAY G.W., 1982 - Metabolic control of fruitbody morphogenesis in *Coprinus cinereus*. *In:* WELLS K. & WELLS E.K., *Basidium and basidiocarps: evolution, cytology, function and development*. New York, Springer Verlag: 157-173.

GRUEN H.E., 1963 - Endogenous growth regulation in carpophores of *Agaricus bisporus*. *Pl. Physiol.* 38: 652-666.

GRUEN H.E., 1967 - Growth regulation in fruiting bodies of *Agaricus bisporus*. *Mushroom Science* 6: 103-120.

GRUEN H.E., 1969 - Growth and rotation of *Flammulina velutipes* fruit bodies and the dependence of stipe elongation on the cap. *Mycologia* 61: 149-166.

GRUEN H.E. and WU S.H., 1972 - Dependence of fruit body elongation on the mycelium in *Flammulina velutipes*. *Mycologia* 64: 995-1007.

GRUEN H.E., 1976 - Promotion of the stipe elongation in *Flammulina velutipes* by a diffusate from excise lamellae supplied with nutrients. *Canad. J. Bot.* 54: 1306 -1315.

GRUEN E.H. and WONG W.M., 1981a - Distribution of cellular amino acids, protein, and total organic nitrogen during fruit-body development in *Flammulina velutipes* I. Growth on sawdust medium. *Canad. J. Bot.* 60: 1330-1341.

GRUEN E.H. and WONG W.M., 1981b - Distribution of cellular amino acids, protein, and total organic nitrogen during fruit-body development in *Flammulina velutipes*. II. Growth on potato-glucose medium. *Canad. J. Bot.* 60: 1342-1351.

GRUEN H.E., 1982 - Control of stipe elongation by the pileus and mycelium in fruitbodies of *Flammulina velutipes* and other agaricales. *In:* WELLS K. & WELLS E.K., *Basidium and basidiocarp: evolution; cytology, function and development*. New York, Springer Verlag: 125-155.

HAGIMOTO H. and KONISHI M., 1959 - Studies on the growth of fruit-body of fungi. I. Existence of a hormone active to the growth of fruit-body in *Agaricus bisporus* (Lange) Sing. *Bot. Mag. (Tokyo)* 72: 359-366.

Source : MNHN, Paris

HAGIMOTO H. and KONISHI M., 1960 - Studies on the growth of fruit-body of fungi. II. Activity and stability of the growth hormone in the fruit-body of *Agaricus bisporus* (Lange) Sing. *Bot. Mag. (Tokyo)* 73: 283-287.

HAGIMOTO H., 1963a - Studies on the growth of fruit-body of fungi. IV. The growth of the fruit-body of *Agaricus bisporus* and the economy of the mushroom growth hormone. *Bot. Mag. (Tokyo)* 76: 256-263.

HAGIMOTO H., 1963b - The mushroom growth hormone and the geotropic response of fruit-body in *Agaricus bisporus*. *Bot. Mag. (Tokyo)* 76: 363-365.

HAMMOND J.B.W. and NICHOLS R., 1976 - Carbohydrate metabolism in *Agaricus bisporus* (Lange) Sing: changes in soluble carbohydrates during growth of mycelium and sporophore. *J. Gen. Microbiol.* 93: 309-320.

HAMMOND J.B.W., 1981 - Variations in enzyme activity during periodic fruiting of *Agaricus bisporus*. *New Phytol.* 89: 419-428.

HEIDE O.M., 1977 - Photoperiod and temperature interactions in growth and flowering of strawberry. *Physiol. Pl.* 40: 21-26.

HORIKOSHI T., KITAMOTO Y. and KASAI Z., 1973 - Distribution of the two respiratory enzymes in developing fruit-bodies of *Favolus arcularius* (Fr.) Ames. *Trans. Mycol. Soc. Japan* 14: 307-309.

HORRIERE F., 1977 - Recherches physiologiques sur la fructification d'un Basidiomycète *Coprinus congregatus* Bull. ex Fr.: influence de quelques facteurs nutritionnels. Thèse Docteur Ingénieur, Université Lyon I.

HUGUENEY R., 1978 - Recherches histologiques et cytologiques sur le développement du carpophore des Coprins; évolution des ultrastructures des articles hyméniaux et de la spore. Thèse Science, Lyon.

INGOLD C.T. and NAWAZ M., 1967 - Sporophore production in *Sphaerobolus* with special reference to periodicity. *Ann. Bot. (London)* 31: 791-802.

KAMADA T., KURITA R. and TAKEMARU T., 1978 - Effects of light on basidiocarp maturation in *Coprinus macrorhizus*. *Pl. Cell Physiol.* 19: 263-275.

KAWAI G. and IKEDA Y., 1982 - Fruiting-inducing activity or cerebrosides observed with *Schizophyllum commune*. *Biochim. Biophys. Acta* 719: 612-618.

KAWAI G., OHNISHI M., FUJINO Y. and IKEDA Y., 1986 - Stimulatory effect of certain sphingolipids on fruiting of *Schizophyllum commune*. *J. Biol. Chem.* 261: 779-784.

KITAMOTO Y. and GRUEN H.E., 1976 - Distribution of cellular carbohydrates during development of the mycelium and fruit-bodies of *Flammulina velutipes*. *Pl. Physiol.* 58: 485-491.

KOMAGATA K. and OKUNISHI M., 1969 - Histochemical studies on the development of carpophores of *Coprinus kimurae*. *J. Gen. Appl. Microbiol.* 15: 231-237.

KÜHNER R., 1926 - Contribution à l'étude des Hyménomycètes et spécialement des Agaricacées. Thèse Sciences, Paris.

KÜHNER R., 1977 - Variation of nuclear behaviour in the homobasidiomycetes. *Trans. Brit. Mycol. Soc.* 68: 1-16.

LEONARD T.J. and DICKS S., 1973 - Induction of haploid fruiting by mechanical injury in *Schizophyllum commune*. *Mycologia* 55: 809-822.

LEONARD T.J. and PHILLIPS L.E., 1973 - Study of phenoloxidase activity during the reproductive cycle of *Schizophyllum commune*. *J. Bacteriol.* 114: 7-10.

LE ROUX P., 1965 - Métabolisme carboné et azoté du champignon cultivé. *Mushroom Science* 6: 170-190.

LESLIE J.F. and LEONARD T.J., 1979 - Monokariotic fruiting in *Schizophyllum commune*. Genetic control of the response to mechanical injury. *Molec. Gen. Genet.* 175: 5-12.

Source : MNHN, Paris

LONG P.E. and JACOBS L., 1974 - Aseptic fruiting of the cultivated mushroom, *Agaricus bisporus*. *Trans. Brit. Mycol. Soc.* 63: 99-107.

LOVETT J.S., 1985 - Fungal differentiation and development: problems and prospects. *In:* TIMBERLAKE W.E., *Molecular genetics of filamentous fungi*. New York, Alan R. Liss Inc.: 187-192.

LU B.C., 1972 - Dark dependence of meiosis at elevated temperatures in the Basidiomycete *Coprinus lagopus*. *J. Bacteriol.* 111: 833-834.

LU B.C., 1982 - Replication of deoxyribonucleic acid and crossing over in *Coprinus*.*In:* WELLS K. & WELLS E.K., *Basidium and basidiocarp: evolution, cytology, function and development*. New York, Springer Verlag: 93-112.

LUKENS R.J., 1966 - Interference of low temperatures with the control of tomato early blight through use of nocturnal illumination. *Phytopathology* 56: 1430-1431.

Mc LAUGHLIN D.J., 1982 - Ultrastructure and cytochemistry of basidial and basidiospores development. *In* WELLS K. & WELLS E.K., *Basidium and basidiocarp: evolution, cytology, function and development*. New York, Springer Verlag: 37-74.

MADELIN M.F., 1956a - Studies on the nutrition of *Coprinus lagopus* Fr. especially as affecting fruiting. *Ann. Bot. (London)* 20: 307-330.

MADELIN M.F., 1956b - The influence of light and temperature on fruiting of *Coprinus lagopus* Fr. in pure culture. *Ann. Bot. (London)* 20: 467-480.

MADELIN M.F., 1960 - Visible changes in the vegetative mycelium of *Coprinus lagopus* Fr. at the time of fruiting. *Trans. Brit. Mycol. Soc.* 43: 105-110.

MANACHERE G., 1961 - Influence des conditions d'éclairement sur la fructification de *Coprinus congregatus* Bull. ex Fr. *Compt. Rend. Hebd. Séances Acad. Sci.*, Sér. D, 252: 2912-2913.

MANACHERE G., 1968 - Influence des conditions d'éclairement sur le déroulement des phénomènes cytologiques au cours de la sporogenèse de *Coprinus congregatus* Bull. ex Fr. *Compt. Rend. Hebd. Séances Acad. Sci.*, Sér. D, 267: 2111-2114.

MANACHERE G., 1970 - Recherches physiologiques sur la fructification de *Coprinus congregatus* Bull. ex Fr.: action de la lumière; rythme de production de carpophores. *Ann. Sci. Nat. Bot.*, Sér. 12, 11: 1-95.

MANACHERE G., 1971 - Research on the fruiting rhythm of a basidiomycete mushroom *Coprinus congregatus* Bull. ex Fr. *J. Interdisciplin. Cycle Res.* 2: 199-209.

MANACHERE G. and ROBERT J.C., 1972 - Fruiting rhythm of a basidiomycete mushroom, *Coprinus congregatus* Bull. ex Fr.: physiological significance of the variation of the rhythm periods in relation to culture temperature. *J. Interdisciplin. Cycle Res.* 3: 135-143.

MANACHERE G., 1974 - *Coprinus congregatus*, un modèle biologique pour l'étude de quelques problèmes généraux posés par la fructification des champignons supérieurs. *Mushroom Science* 9: 783-798.

MANACHERE G., 1976 - Substance(s) produite(s) par les carpophores mûrs de *Coprinus congregatus* et régulatrice(s) du rythme de fructification du champignon; délais nécessaires à leur libération. *Rev. Mycol. (Paris)* 40: 329-339.

MANACHERE G., 1977 - Modifications du rythme de fructification de *Coprinus congregatus* par le dépôt de carpophores mûrs exogènes à la surface des cultures. *Physiol. Pl.* 39: 201-205.

MANACHERE G., 1978 - Morphogenèse des carpophores de basidiomycètes supérieurs. Connaissances actuelles. *Rev. Mycol. (Paris)* 42: 191-252.

MANACHERE G., 1980 - Conditions essential for controlled fruiting of macromycetes. A review. *Trans. Brit. Mycol. Soc.* 75: 255-270.

MANACHERE G. and BASTOUILL - DESCOLLONGES Y., 1982 - Recherches cytophysiologiques sur la sporogenèse de *Coprinus congregatus* Bull. ex Fr.: introduction à

Source : MNHN, Paris

l'étude du déroulement de la méiose en rapport avec les conditions lumineuses et thermiques. *Cryptogamie, Mycol.* 3: 391-408.

MANACHERE G. and BASTOUILL - DESCOLLONGES Y., 1983 - Effect of "reduced" photoperiods on development of carpophores of *Coprinus congregatus*. *Trans. Brit. Mycol. Soc.* 81: 630-633.

MANACHERE G., ROBERT J.C., DURAND R., BRET J.P. and FEVRE M., 1983 - Differentiation in the basidiomycetes. *In:* SMITH J.E., *Fungal differentiation. a contemporary synthesis*. New York, Marcel Dekker Inc.: 481-514.

MANACHERE G., 1985 - Sporophore differentiation of higher fungi: a survey of some actual problems. *Physiol. Vég.* 23: 221-230.

MANACHERE G.and BASTOUILL - DESCOLLONGES Y., 1985 - An analysis of photo-induced fruiting in *Coprinus congregatus*. *Photochem. & Photobiol.* 42: 725-729.

MATTHEWS T.R. and NIEDERPRUEM D.J., 1973 - Differentiation in *Coprinus lagopus* II. Histology and ultrastructural aspects of developing primordia. *Arch. Mikrobiol.* 88: 169-180.

MEINHARDT F. and ESSER K., 1983 - Genetic aspects of sexual differentiation in fungi. *In:* SMITH J.E., *Fungal differentiation. a contemporary synthesis*. New York, Marcel Dekker Inc.: 537-557.

MIYAKE H., TANAKA K. and ISHIKAWA T., 1980 - Basidiospore formation in monokaryotic fruiting bodies of a mutant strain of *Coprinus macrorhizus*. *Arch. Microbiol.* 126: 207-212.

MOORE D. and AL GHARAWI A., 1976 - An elevated level of NADP-linked glutamate dehydrogenase is not a general feature of the caps of agaric sporophores. *Trans. Brit. Mycol. Soc.* 66: 149-150.

MOORE D. and EWAZE J.O., 1976 - Activities of some enzymes involved in metabolism of carbohydrate during sporophore development in *Coprinus cinereus*. *J. Gen. Microbiol.* 97: 313-322.

MOORE D., FLEITI M.Y. and BUTLER R.D., 1979 - Morphogenesis of the carpophore of *Coprinus cinereus*. *New Phytol.* 83: 695-722.

MOORE D., 1984a - Review - Developmental biology of the *Coprinus cinereus* carpophore: metabolic regulation in relation to cap morphogenesis. *Exp. Mycol.* 8: 283-297.

MOORE D., 1984b - Positional control of development in fungi. *In:* BARLOW P.W. & CARR D.J., *Positional controls in plant development*. Cambridge University Press: 107-135.

MOORE D., CASSELTON L.A., WOOD D.A. and FRANKLAND J.C., 1985 - *Developmental biology of higher fungi*. Cambridge University Press, 515 p.

MOORE D., LIU M. and KUHAD R.C., 1987 - Karyogamy-dependent enzyme derepression in the basidiomycete, *Coprinus*. *Cell Biol. Intern. Rep.* 11: 335-341.

MORIMOTO N., SUDA S. and SAGARA N., 1981 - Effect of ammonia on fruit-body induction of *Coprinus cinereus* in darkness. *Pl. Cell Physiol.* 22: 247-254.

MULDER G.H. and WESSELS J.G.H., 1986 - Molecular cloning of RNAs differentially expressed in monokaryons and dikaryons of *Schizophyllum commune* in relation to fruiting. *Exp. Mycol.* 10: 214-227.

MURAO S., HAYASHI H. and TARUI N., 1984 - Anthranilic acid, as a fruiting body inducing substance in *Favolus arcularius*, from a strain TA 7 of Actinomycetes. *Agric. Biol. Chem.* 48: 1669-1671.

NETZER U. von, 1977 - A system for the investigation of morphogenesis in Basidiomycetes. 2nd International Mycological Congress, Tampa (Florida). Abstracts: 474.

OKUNISHI M. and KOMAGATA K., 1975 - Histochemical studies on the development of carpophore of *Polyporus brumalis* (Pers. ex Fr.) Karst. *J. Gen. Appl. Microbiol.* 21: 211-216.

Source : MNHN, Paris

PLUNKETT B.E., 1958 - Translocation and pileus formation in *Polyporus brumalis*. *Ann. Bot. (London)* 22: 237-249.

RAO P.S. and NIEDERPRUEM D.J., 1969 - Carbohydrate metabolism during morphogenesis of *Coprinus lagopus* (sensu Buller). *J. Bacteriol.* 100: 1222-1228.

RAPER C.A., 1983 - Control for development and differentiation of the dikaryon in Basidiomycetes. *In:* BENETT J.W. & CIEGLER A., *Secondary metabolism and differentiation in fungi*. New York, Marcel Dekker Inc.: 195-238.

RAPER J.R. and KRONGELB G.S., 1958 - Genetic and environmental aspects of fruiting in *Schizophyllum commune* Fr. *Mycologia* 50: 707-740.

RAUDASKOSKI M. and LU B.C., 1980 - The effect of hydroxyurea on meiosis and genetic recombination in the fungus *Coprinus lagopus*. *Canad. J. Genet. Cytol.* 22: 41-50.

RAUDASKOSKI M. and SALONEN M., 1984 - Interrelationships between development and basidiocarp initiation. *In:* JENNINGS D.H. & RAYNER A.D.M., *The ecology and physiology of the fungal mycelium*. Cambridge University Press: 291-322.

RAUDASKOSKI M. and YLI-MATTILA T., 1985 - Capacity for photoinduced fruiting in a dikaryon of *Schizophyllum commune*. *Trans. Brit. Mycol. Soc.* 85: 145-151.

REIJNDERS A.F.M., 1983 - *Les problèmes du développement des carpophores des Agaricales et de quelques groupes voisins*. The Hague, W. Junk, 412 p.

REIJNDERS A.F.M. and MOORE D., 1985 - Developmental biology of agarics - An overview. *In:* MOORE D., CASSELTON L.A., WOOD D.A. & FRANCKLAND J.C., *Developmental biology of higher fungi*. Cambridge University Press: 581-595.

ROBERT J.C., 1971 - Effect favorable d'une période froide sur la maturation de carpophores de *Coprinus congregatus* Bull. ex Fr. inhibés par un éclairement continu. *Compt. Rend. Hebd. Séances Acad. Sci.*, Sér. D, 273: 154-157.

ROBERT J.C. and MANACHERE G., 1971 - Influence de la température sur le rythme de fructification de *Coprinus congregatus* Bull. ex Fr.: remarques préliminaires. *Compt. Rend. 96è Congr. Natl. Soc.Sav. (Toulouse, Avril 1971), Sect. Sci.*, 3: 119-129.

ROBERT J.C., 1977a - Fruiting of *Coprinus congregatus*: biochemical changes in fruitbodies during morphogenesis. *Trans. Brit. Mycol. Soc.* 68: 379-387.

ROBERT J.C., 1977b - Fruiting of *Coprinus congregatus*: relationship to biochemical changes in the whole culture. *Trans. Brit. Mycol. Soc.* 68: 389-395.

ROBERT J.C., 1978 - Quelques aspects métaboliques de la fructification du basidiomycète *Coprinus congregatus* Bull. ex Fr. *Mushroom Science* 10: 583-702.

ROBERT J.C. and DURAND R., 1979 - Light and temperature requirements during fruit body development of a basidiomycete mushroom, *Coprinus congregatus*. *Physiol. Pl.* 46: 174-178.

ROBERT J.C., 1982 - Corrélations physiologiques régularisant la morphogenèse des carpophores de *Coprinus congregatus* Bull. ex Fr.: influence des facteurs externes sur divers aspects métaboliques; introduction à la connsaissance des mécanismes du rythme de fructification. Thèse Doctorat d'état, Université Lyon I, Villeurbanne.

ROBERT J.C. and BRET J.P., 1987 - Release of an inhibitor of stipe elongation from illuminated caps of *Coprinus congregatus*. *Canad. J. Bot.* 65: 505-508.

ROSIN I.V. and MOORE D., 1985 a - Origin of the hymenophore and establishment of major tissue domains during fruit body development in *Coprinus cinereus*. *Trans. Brit. Mycol. Soc.* 84: 609-619.

ROSIN I.V. and MOORE D., 1985 b - Differentiation of the hymenium in *Coprinus cinereus*. *Trans. Brit. Mycol. Soc.* 84: 621-624.

ROSS I.K., 1982 - Localization of carpophore initiation in *Coprinus congregatus*. *J. Gen. Microbiol.* 128: 2755-2762.

Source : MNHN, Paris

ROSS I.K., 1985 - Determination of the initial steps in differentiation in *Coprinus congregatus*. *In:* MOORE D., CASSELTON L.A., WOOD D.A. & FRANKLAND J.C., *Developmental biology of higher fungi*. Cambridge University Press: 353-373.

RUSMIN S. and LEONARD T.J., 1975 - Biochemical induction of fruiting bodies in *Schizophyllum commune*: a bioassay and its application. *J. Gen. Microbiol.* 90: 217-227.

RUSMIN S. and LEONARD T.J., 1978 - Biochemical induction of fruiting in *Schizophyllum commune*. Isolation and preliminary purification of an inducing substances from *Agaricus bisporus* mushrooms. *Pl. Physiol.* 61: 538-543.

SALAS J.A. and HANCOCK J.G., 1972 - Production of the perfect stage of *Mycena citricolor* (Berk. and Curt.) Sacc. *Hilgardia* 41: 213-234.

SCHWALB M.N., 1974 -Effects of adenosine 3'-5'-cyclic monophosphate on the morphogenesis of fruit-bodies of *Schizophyllum commune*. *Arch. Mikrobiol.* 96: 17-20.

SCHWALB M.N. and MILES P.G., 1978 - *Genetics and morphogenesis in the basidiomycetes*. New York, Academic Press, 168 p.

SMITH J.E., 1983 - *Fungal differentiation: a contemporary synthesis*. New York, Marcel Dekker Inc., 624 p.

STAHL U. and ESSER K., 1976 - Genetics of fruit-body production in higher basidiomycetes. I. Monokaryotic fruiting and its correlation with dikaryotic fruiting in *Polyporus ciliatus*. *Molec. Gen. Genet.* 148: 183-197.

STEWART G.R. and MOORE D., 1974 - The activity of glutamate dehydrogenases during mycelial growth and sporophore development in *Coprinus lagopus* (sensu Lewis). *J. Gen. Microbiol.* 83: 73-81.

THIELKE C.T., 1982 - Meiotic divisions in the basidium. *In:* WELLS K. & WELLS E.K., *Basidium and basidiocarp: evolution, cytology, function and development*. New York, Springer Verlag: 75-91.

TIMBERLAKE W.E., 1985 - *Molecular genetics of filamentous fungi*. New York, Alan R. Liss Inc., 465 p.

TURIAN G., 1969 - *Différenciation Fongique*. Paris, Masson.

TURIAN G., 1983 - Concept of fungal differentiation. *In:* SMITH J.E., *Fungal differentiation: a contemporary synthesis*. New York, Marcel Dekker Inc.: 1-18.

TURNER E.M., 1974 - Phenoloxidase activity in relation to substrate and development stage in the mushroom, *Agaricus bisporus*. *Trans. Brit. Mycol. Soc.* 63: 541-547.

TURNER E.M., 1977 - Development of excised sporocarps of *Agaricus bisporus* and its control by CO2. *Trans. Brit. Mycol. Soc.* 69: 183-186.

UNO I. and ISHIKAWA T., 1971 - Chemical and genetical control of induction of monokaryotic fruiting-bodies in *Coprinus macrorhizus*. *Molec. Gen. Genet.* 113: 228-239.

UNO I. and ISHIKAWA T., 1973 - Purification and identification of the fruiting inducing substances in *Coprinus macrorhizus*. *J. Bacteriol.* 113: 1240-1248.

UNO I. and ISHIKAWA T., 1976 - Effect of cyclic AMP on glycogen phosphorylase in *Coprinus macrorhizus*. *Biochim. Biophys. Acta* 452: 112-120.

UNO I. and ISHIKAWA T., 1982 - Biochemical and genetic studies on the initial events of fruit-body formation. *In:* WELLS K. & WELLS E.K., *Basidium and basidiocarp: evolution, cytology, function and development*. New York, Springer Verlag: 113-123.

VERRINDER GIBBINS A.M. and LU B.C., 1984 - Induction of normal fruiting on originally monokaryotic cultures of *Coprinus cinereus*. *Trans. Brit. Mycol. Soc.* 83: 331-335.

VIOT M., 1970 - Mise en évidence de corrélations de croissance au cours du développement des carpophores d' *Asterophora parasitica* (Bull. ex Fr.) Sing. *Compt. Rend. Hebd. Séances Acad. Sci.*, Sér. D, 270: 790-792.

Source : MNHN, Paris

WELLS K. and WELLS E.K., 1982 - *Basidium and basidiocarp: evolution, cytology, function and development.* New York, Springer Verlag, 187 p.

WESSELS J.G.H., 1965 - Morphogenesis and biochemical processes in *Schizophyllum commune* Fr. *Wentia* 13: 1-113.

WESSELS J.G.H., 1978 - Incompatibility factors and the control of biochemical processes. *In:* SCHWALB M.N. & MILES P.G., *Genetics and morphogenesis in the Basidiomycetes.* London, New York, Academic Press: 81-104.

WESSELS J.G.H., 1985 - Gene expression during basidiocarp formation in *Schizophyllum commune*. *In:* TIMBERLAKE W.E., *Molecular genetics of filamentous fungi.* New York, Alan R. Liss Inc.: 193-206.

WILKINS M.B., 1969 - *Physiology of plant growth and development.* London, MacGraw-Hill, 695 p.

WOOD D.A., 1976 - Primordium formation in axenic cultures of *Agaricus bisporus* (Lange) Sing. *J. Gen. Microbiol.* 95: 313-323.

WOOD D.A. and GOODENOUGH P.W., 1977 - Fruiting of *Agaricus bisporus*. Changes in extracellular enzyme activities during growth and fruiting. *Arch. Microbiol.* 114: 161-165.

WOOD D.A., 1979 - Studies on primordium initiation in *Agaricus bisporus* and *Agaricus bitorquis* (syn. *edulis*). *Mushroom Science* 10: 565-586.

YLI-MATTILA T., 1985 - Action spectrum for fruiting in the basidiomycete *Schizophyllum commune*. *Physiol. Pl.* 65: 287-293.

YLI-MATTILA T., 1987 - The effect of UV-A light on cAMP level in the basidiomycete *Schizophyllum commune*. *Physiol. Pl.* 69: 451-455.

Source : MNHN, Paris